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A biopsychosocial approach to the examination of potential biomarkers of depression, anxiety and wellbeing in adolescence.

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Declaration

I hereby declare that this thesis:

a) Has been composed entirely by myself
b) Is my own original work
c) Has not been submitted for any other degree or professional qualification

Eilidh Smith

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Abstract

Adolescence constitutes a period of vulnerability for the onset of psychological distress. The onset of psychological illness during this period is associated with a more severe and enduring course. Considerable research investigates risk factors and precursors of illness but is largely dependent on research involving adult samples. Additionally, the majority of research focuses on symptoms of distress without examining measures of wellbeing. This is a significant limitation as it may only partly capture experiences. This thesis will examine symptoms of depression and anxiety related distress alongside measures of wellbeing in adolescence.

Cognitive biases, attachment, emotional reactivity, emotional regulation, the stress-response system and sleep are all components that have been implicated by previous research as related to the onset of disorders; but, lack robust findings within adolescent samples. This thesis comprises of four studies which discuss empirical research examining these factor and the extent to which they are able to predict symptoms of depression, anxiety and wellbeing. Two different samples were involved in this research. Data informing chapters Two and Three are based on the same sample, while data informing chapters Five and Six are from a second sample. Both samples comprise of adolescents aged 13-18, who were recruited from Scottish secondary schools and participated in self-report measures, experimental paradigms and provided biological samples for analysis.

Overall, results suggest that tasks assessing self-referential memory and interpretation of ambiguous scenarios, alongside measures of rumination and dysfunctional attitudes, were able to predict symptoms of depression, anxiety and wellbeing. Regression models were able to predict approximately 60% of the variance in depression, anxiety and wellbeing.

Activation of the stress-response system was approximated via saliva and hair samples analysed for cortisol exposure. Results indicate that physiological measures were shown to have less predictive utility than cognitive mechanisms. Analyses indicate that lowered morning and higher evening cortisol was associated with increased depression and lower wellbeing. However, although this relationship was significant, it was small and may have limited clinical utility. Three-month retrospective measurement of cortisol was not significantly related to measures of depression, anxiety or wellbeing.

The examination of sleep architecture, using measured through Consensus Sleep Diaries and *Philips Actiwatch 2* actigraphic wrist monitors partially supported relationships between depression and sleep. Self-report measures were significantly predictive of depression, anxiety

and wellbeing. Specifically, increased wakefulness while in bed is implicated in increased depression and anxiety and lower wellbeing.

Overall results indicate that psychological factors are more salient markers of mental health than physiological markers in this sample. Overall results emphasise the importance of psychological factors suggesting that these may be ideal targets for developmentally and individually appropriate interventions. These results have implications for the identification of individuals at risk for onset of mental health conditions as well as psychological components of importance for wellbeing.

Lay Summary

Mental health can be understood as a spectrum that runs from health to ill-health. Depression, anxiety and wellbeing are all parts of this spectrum. This thesis aims to examine factors that may be important for maintaining wellbeing and reducing depression and anxiety. It is particularly important to look at these factors in adolescents because adolescence is the period at which depression and anxiety originate for a large proportion of people. Adolescents may be affected by risk factors in a different way to adults and this could be an age that intervention to improve mental health is more successful. Neuroticism is a personality trait that is thought to increase the risk for both depression and anxiety. Neuroticism predicts depression and anxiety to some extent. This thesis aims to measure other factors that may account for symptoms of depression, anxiety and wellbeing that are not accounted for by neuroticism in adolescents.

Factors that contribute to depression and anxiety are not fully understood. Previous research has found that people with depression and anxiety process information in a more negative way than people who are healthy. Results of this thesis show that the extent to which adolescents identify and recall more negative information and interpret ambiguous situations as negative explains more symptoms of depression, anxiety and wellbeing than neuroticism alone. This suggests that these factors may contribute to psychological health and could be targeted by treatment to improve mental health.

Another factor that may be involved in shaping mental health is the stress-response system. This is a physiological system that helps us react to situations that might be dangerous. Measuring the levels of the hormone cortisol is a way to estimate how activated the stress-response system is. If this system is activated too frequently it is potentially harmful. Some research has suggested that mental health disorders are related to changes to the way this system is activated. This thesis did not strongly support this theory, as measures of cortisol were not strongly related to depression, anxiety or wellbeing. However, the way adolescents experience emotions was related to the stress-response system, this could be important for understanding the relationship between mental and physical health as well as identifying a potential target for treatment.

Problems with sleep are very common in people that have mental health difficulties and have been shown to be important for wellbeing. Sleep can be measured subjectively and objectively. This thesis has combined subjective and objective measurements to understand more about

the role of sleep in depression, anxiety and wellbeing. It was shown that subjective measures were more strongly related to depression, anxiety and wellbeing than objective measures. This might suggest that the way individuals interpret the way they have slept is strongly related to their mental health, but that this may not be a very accurate representation of objectively measured components of sleep. This may be useful in helping to recognise people who might be at increased risk of experiencing higher levels of depression and anxiety and lower levels of wellbeing. In turn trying to improve sleep may protect against depression and anxiety and promote wellbeing.

This thesis has also implicated the level of security they feel in relationships with others as important for understanding depression, anxiety and wellbeing. The implications of this thesis are that focusing on cognitive components and sleep might be helpful for improving the mental health of adolescents and important for preventing illness. Finally, the implications of results, limitations of this work and ideas for future research are discussed.

Contents

List of Abbreviations

ABMT	Autobiographical Memory Task
AMHS	Adult Mental Health Services
AOR	Adjusted Odds Ratio
APA	American Psychiatric Association
AST	Ambiguous Scenarios Task
AuC	Area Under the Curve
AuCg	Area Under the Curve with respect to ground
AuCi	Area Under the Curve with respect to increase
BBC	BBC subjective Wellbeing Scale
BMI	Body Mass Index
CAMHS	Child and Adolescent Mental Health Services
CAR	Cortisol Awakening Response
CBT	Cognitive Behavioural Therapy
CCDS	Core Consensus Sleep Diary
CRH	Corticotropin-Releasing Hormone
DAS	Dysfunctional Attitudes Scale
DSM	Diagnostic and Statistical Manual
ELISA	Enzyme-Linked Immunosorbent Assay
EPQ	Eysenck Personality Questionnaire
ERQ	Emotional Regulation Questionnaire
ERS	Emotional Reactivity Scale
FDR	False Discovery Rate
GP	General Practitioner
HPA	Hypothalamic Pituitary Adrenal
ICD	International Classification of Diseases
ID	Identification
IPPA	Inventory of Parent and Peer Attachment
LC-MS/MS	Liquid Chromatography-Mass Spectrometry
MDD	Major Depressive Disorder
MFQ	Mood and Feelings Questionnaire
MRI	Magnetic Resonance Imaging
NHS	National Health Service
NICE	National Institute of Clinical Excellence
PSG	polysomnography
RRS	Ruminative Response Scale
SAMH	Scottish Association for Mental Health
SCAS	Spences Children's Anxiety Scale
SLE	Stressful Life Events
SOL	Sleep Onset Latency
SRR	Self-Referential Recall

SRRT	Self-Referential Recall Task
SSRI	Selective Serotonin Reuptake Inhibitor
TADS	Treatment of Adolescent Depression Trial
TST	Total Sleep Time
VIF	Variance Inflation Factor
WASO	Wake After Sleep Onset
WHO	World Health Organisation

Publications Arising From This Work

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Chapter One - Depression, the spectrum of mental health, and adolescent development

The aim of this chapter is to outline the concepts that are the main focus of this thesis: the spectrum of depression and wellbeing, the adolescent period, and the relationship between these two components. Throughout this introduction theoretical positions, current health policies and epidemiology will be introduced. These topics will be continued in subsequent chapters where relevant. This introductory chapter will also explore the conceptualisation of neuroticism, which has been considered a means of indexing risk for onset of disorder and is measured throughout these studies. Cognitive and biological risk factors will be introduced briefly within this chapter and will be considered more fully within subsequent chapters.

1.1 Defining depression

Depression is a severely disabling disorder, characterised by low mood or anhedonia, and is the leading cause of disability worldwide (World Health Organisation, 2017). Depression refers to a group of disorders including dysthymia, major depressive disorder, and premenstrual dysthymic disorder (APA, 2013). After classification of specific disorder type, depressive symptoms can be categorised as mild, moderate or severe, and as current, partially or fully remitted. Literature, and this thesis, consider depression to relate to major depressive disorder (henceforth 'depression') and/or its symptoms. Within research, diagnostic and symptom measures are typically based on the criteria defined by the American Psychiatric Association's (APA's) Diagnostic and Statistical Manual of Mental Disorders (DSM-5; APA, 2013). Diagnostic criteria of depression are based on the experience of depressed mood or loss of interest or pleasure in addition to at least four of the following symptoms: significant changes in weight, appetite, sleep, activity, feelings of guilt or worthlessness, fatigue or loss of energy, diminished concentration, suicidality or recurrent thoughts of death, for at least a two-week period which represents a change from prior functioning and causes clinically significant distress or impairment in social or occupational functioning (APA, 2013, see Figure 1.1). These symptoms must not be otherwise explained by substance use or misuse or another medical condition (APA, 2013). The presence of symptoms varies between individuals, loss

of pleasure or interest is particularly prominent, and patients may initially present with somatic complaints or fatigue (APA, 2013).

Major Depressive Disorder (DSM-5, 2013)

A. Five (or more) of the following symptoms have been present during the same 2-week period and represent a change from previous functioning; at least one of the symptoms is either (1) depressed mood or (2) loss of interest or pleasure.
Note: Do not include symptoms that are clearly attributable to another medical condition.

1. Depressed mood most of the day, nearly every day, as indicated by either subjective report (e.g. feels sad, empty hopeless) or observation made by others (e.g. appears tearful). (***Note:*** In children and adolescents, can be irritable mood.)
2. Markedly diminished interest or pleasure in all, or almost all, activities most of the day, nearly every day (as indicated by either subjective account or observation).
3. Significant weight loss when not dieting or weight gain (e.g. a change of more than 5% of body weight in a month), or decrease or increase in appetite nearly every day.
   (***Note:*** In children, consider failure to make expected weight gain.)
4. Insomnia or hypersomnia nearly every day.
5. Psychomotor agitation or retardation nearly every day (observable by others, not merely subjective feelings of restlessness or being slowed down.)
6. Fatigue or loss of energy nearly every day.
7. Feelings of worthlessness or excessive or inappropriate guilt (which may be delusional) nearly every day (not merely self-reproach or guilt about being sick).
8. Diminished ability to think or concentrate, or indecisiveness, nearly every day. (Either by subjective account or as observed by others).
9. Recurrent thoughts of death (not just fear or dying), recurrent suicidal ideation without a specific plan, or a suicide attempt, or a specific plan for committing suicide.

B. The symptoms cause clinically significant distress or impairment in social, occupational, or other important areas of functioning.

C. The episode is not attributable to the physiological effects of a substance or to another medical condition.

Figure 1.1 DSM-5 (APA, 2013) Criteria for Major Depressive Disorder

Depression is often comorbid with other conditions, including physical conditions and other mental health disorder and has been associated with poorer healthcare outcomes of chronic conditions such as asthma, diabetes and arthritis (Moussavi et al., 2007). Other psychiatric and personality disorders, such as substance abuse (Swendsen and Merikangas, 2000) and impulse control disorder (Kessler, Berglund, Demler et al., 2003) have also been associated. Additionally, depression is highly comorbid with other psychiatric diagnoses. Kessler et al., (2003) demonstrated that 72% of individuals meeting the criteria for lifetime depression also met the criteria for at least one other psychiatric disorder. Anxiety and depression are

particularly comorbid disorders, Hirschfield (2001) indicates that anxiety disorders are present in approximately half of patients in the community with depression; this figure rises to 75% if patients recruited from primary care settings are considered. Within the context of children and adolescents, depression was found to be associated with other disorders in 64% of cases. Adolescents (aged 13-18 years) were four times as likely to have a comorbid depression and anxiety disorder (OR 3.96, Avenevoli, Swendsen He, Burstein and Merikangas, 2015).

1.2 Adolescence: a period of increased vulnerability to psychological distress

The term 'adolescence' is often used to refer to the period between childhood and adulthood and is particularly associated with teenage years and puberty (Sawyer et al., 2012). Within the literature these terms (adolescence, young people, teenagers, etc.) are often applied interchangeably with a lack of clarity as to their precise definition. Culturally, this term varies and is often applied to those still in school or secondary education. In practical terms, National Institute of Clinical Excellence (NICE) guidelines (2005) consider Child and Adolescent Mental Health Services (CAMHS) to be available for children and adolescents aged up to 18 years that are experiencing mental health concerns, after which young people would be referred or transferred to Adult Mental Health Services. An increase in understanding of the significance of mental health problems in young people, improved understanding of neurological development, and concerns regarding the success of transitions (or consequences of poor transitions) to adult services has resulted in a desire for CAMHS to treat young people up to the age of 25 (e.g. The Young People's Health Partnership and the Youth Access, see Policy Briefing, 2017).

Recent views regarding the adolescent period support increasing the upper age limit to as high as the mid-twenties due to ongoing neurological development (Casey et al., 2008). Nevertheless, for the purpose of this thesis, the term 'adolescence' will be applied throughout and will refer generally to young people aged 12-18 years. This is in order to directly relate previous literature to the empirical studies conducted (see Chapters Three, Four and Five), as these depend upon individuals recruited from secondary school settings. This has been considered as of particular importance due to the impact of social development and the distinct peer environment of this developmental stage. Furthermore, research has demonstrated that this is the age at which the onset of psychiatric disorders sharply increases (Kessler et al., 2005; Jaworska and MacQueen, 2015). Within the literature, studies of adolescence typically

include youth aged 12-18 years (Jaworska and MacQueen 2015; March et al., 2004), hence this formed the basis of our inclusion criteria during recruitment.

Adolescence is a key stage for the development of emotional disorders. Prevalence estimates vary within this age range with lifetime prevalence of major depressive disorder (MDD) in adolescents (13-18-year-olds) cited at around 11% (Avenevoli et al., 2015; see Figure 1.2). However, estimates are not consistent within this age group. A meta-analysis of epidemiological studies demonstrated an overall prevalence estimate of 5.6% for 13-18-year-olds (Costello, Erkanli and Angold, 2006); although prevalence estimates are as high as 28% of those under 19 (Lewinsohn et al., 1998). This may be partially attributable to the variation of age range within samples. The NICE guidelines (2015) recognise the increase in prevalence of depression following puberty and consider hormonal changes, cognitive maturity, changes in social support and increased life stress to be key contributory factors. Notably, diagnosis of depressive illness in this age group may be complicated by the typical variability of mood in this developmental stage (NICE Guidelines, 2015). Furthermore, the NICE Guidelines (2015) stress a lack of reporting of symptoms by adolescents and an under recognition of symptoms by parents and/or guardians.

The presentation of adolescent depression is similar to that in adults but encompasses distinctive features. DSM-5 (APA, 2013) highlights that in children and adolescents, presentation of depression may differ from adults and that adolescents may display the following symptoms: vague, non-specific physical complaints; absence from school or poor performance in school; being bored; alcohol or substance use; increased irritability, anger or hostility; and reckless behaviour. The critical criterion of diagnosis, impairment of functioning, is of particular relevance in the adolescent context, as this is an educational stage where underperforming academically may have far-reaching consequences. Fergusson, Boden and Horwood (2007) demonstrated that depression at ages 16-21 was associated with increased association with further adverse mental health outcomes, but also lower rates of degree and tertiary education attainment, unemployment and lower income at age 25 years (Fergusson et al., 2007; Dunn and Goodyer, 2006).

While adolescence is a period of heightened vulnerability for depressive disorders, anxiety disorders (including generalized anxiety, social anxiety, panic disorder) also demonstrate increased occurrence during this developmental stage. Similar to depression, anxiety can demonstrate a chronic and recurrent course (Fisak, Richard and Mann, 2011). Depression and

anxiety frequently cooccur, have bidirectional associations and are considered risk factors for further psychopathology. Consequently, depression and anxiety are often examined simultaneously and are frequently referred to as the 'internalising disorders'. Prior research indicates that comorbid anxiety and depression are associated with greater impairment (such as academic difficulties, health problems and suicide attempts) and severity of the primary diagnosis (see Cummings, Caporino and Kendall, 2014 for review). Additionally, anxiety and depressive disorders have been shown to share genetic risk factors (e.g. Hettema, 2008).

Many of the major psychiatric disorders, including depression, are hypothesised to have origins in adolescence (MQ, 2016). Importantly, earlier onset of depression is predictive of longer episodes, a more severe course, poorer recovery and higher rates of recurrence (Dunn and Goodyer; 2006). Depressive episodes in adolescence have been associated with a broad variety of mental health, educational and economic outcomes. This evidence speaks strongly to the economic imperative for early and effective intervention in order to reduce both economic and healthcare burdens.

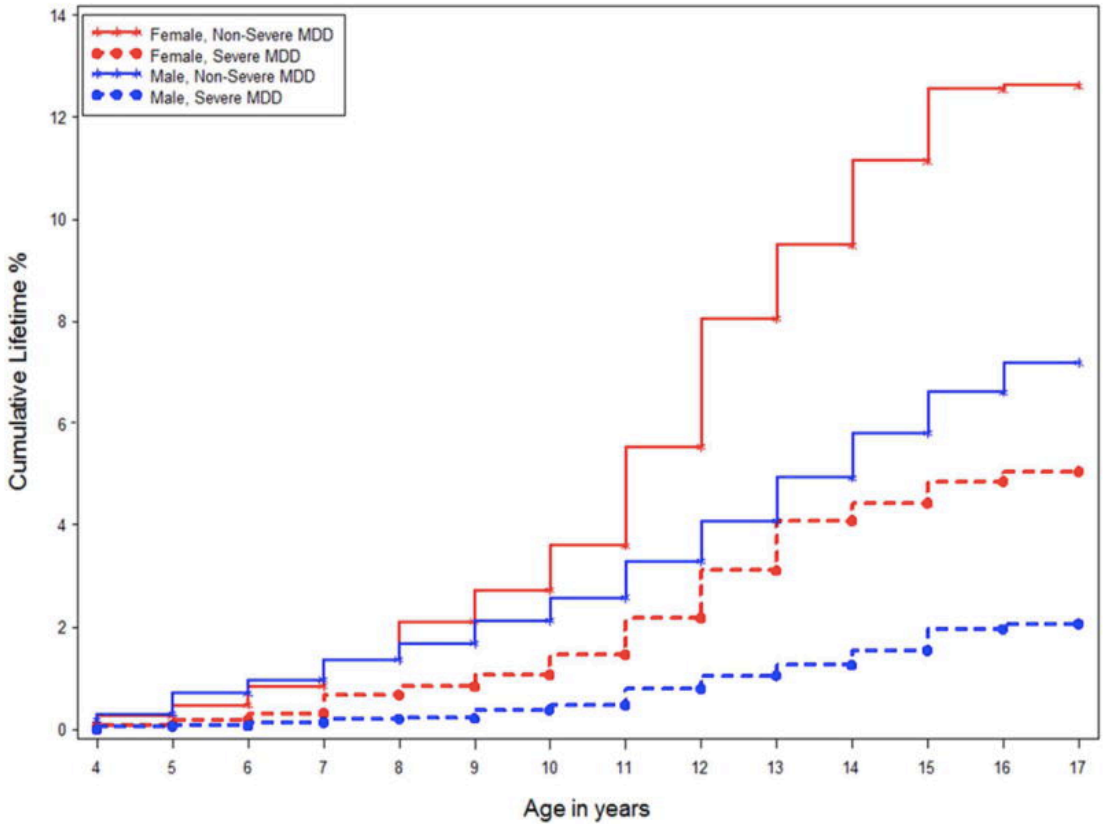


Figure 1.2: Avenevoli et al., (2015) Cumulative lifetime prevalence of major depressive disorder my sex and severity

Neurobiological development occurring in adolescence has been implicated as a potential explanation for the sharp increase of disorder onset in this age group. For example, the prefrontal cortex, which is associated with the top-down regulation of behaviour and emotions, undergoes the most pronounced and protracted development during this period (Sawyer et al., 2012; Choudhury, Blakemore, Charman 2006). A longitudinal MRI study consisting of 145 participants (each participant had between 1-5 scans; 98 participants had at least two scans), demonstrated increasing volume of white matter and decreasing volume of frontal grey matter across adolescence (Giedd et al., 1999). Of note is that, significant structural development of the prefrontal cortex was indicated from approximately 11 to 20 years of age (Giedd et al., 1999). Consequently, this brain region may be relatively underdeveloped in comparison to other neural areas including emotional processing regions such as the limbic system (Casey, Jones, Hare 2008). The implication of this is that a maturational mismatch between cognitive and emotional brain regions exists, which may, to a certain extent, explain increased emotional reactivity in adolescence and the onset of disorders in this age group (Casey et al., 2008).

Research indicates ongoing brain development across the adolescent period continuing into adulthood. For example, adolescents demonstrate increased connectivity between distinct brain regions and greater efficiency and increased processing of resources. This is partly due to changes in the brains axons, which become more insulated by myelin, and dendrites which grow more branches thereby increasing connectivity. This contributes to a linear increase in white matter (Foulkes and Blakemore, 2018). Cortical grey matter volume has been found to decrease in volume and thickness in frontal, parietal and temporal corticies during adolescence, until the early twenties. This is partly attributable to synaptic pruning (see Griffin, 2017 and Foulkes and Blakemore, 2018 for review). This serves to enhance and 'fine tune' functional capacity (Gidd et al., 1999; see Miguel-Hidalgo, 2013 for review). This allows for modification in response to environmental, social, emotional and behavioural stimuli. Such age-related neurobehavioural plasticity of the maturing brain may allow for enhanced ability to process complex information, develop skills and solve problems. However, this age also reflects increased vulnerability whereby social, biological, emotional and psychological factors may have a significant negative impact which has the potential to result in structural or functional alterations leading to lifelong susceptibility to mental ill-health (e.g. Steinberg, 2005; see also review by Griffin, 2017). Demographic and environmental influences have been related to impact neurocognitive development in adolescents, which may have significant long-term effects. For example, individual differences in perceived social status was associated with increased activity in the medial prefrontal cortex, praecuneus and left posterior

superior temporal sulcus (see Foulkes and Blakemore, 2018). As functional fine tuning in adolescence is related to the frequency of activation of particular pathways, individual differences in brain activity may exert long lasting changes. The rapid change in adolescents physically, neurologically and socially seems to present vulnerability to the onset of depression. However, this may also reflect a window of opportunity for the prevention or treatment of disorders that may offer the possibility of developing early intervention or prevention strategies with the potential for lifelong impact.

For example, myelination has been demonstrated to occur within fronto-temporal cortical areas into late-adulthood. Difficulty exists in defining this developmental period due to conflicting results and that different brain areas seem to develop at varying ages. Casey et al., (2008) demonstrated an initial surge of neuronal growth prior to the onset of puberty resulting in a thickening of grey matter. Following this, it is thought that dendritic pruning and myelination, similar to that which occurs during infancy,

Mental Health and Ill-Health

Depression as a Spectrum

The use of diagnostic criteria in the diagnosis of depression as a unitary construct based on exceeding threshold criteria has been under criticism over recent years (Kinderman and Cooke, 2017). This is due to the variability of symptoms experienced by individuals. Kinderman and Cooke (2017) argue that previous research has demonstrated that symptom presentation does not accurately map onto the clinical diagnoses that exist within the manual. This causes significant problems for clinicians as treatment is based on symptom presentation and 'disorder' classification. Kinderman and Cooke (2017) strongly advocate a move away from the current top-down approach to diagnosis as the criteria for classification results in the consideration of individuals' problems as pathological thereby overlooking important social nature and causes facilitating inappropriate management. Furthermore, the pathologising of symptoms or behaviours ignores the reality that distress can exist as a normal part of human experience and drives the need for diagnoses in order to validate symptoms and access treatment (Kinderman and Cooke, 2017). Depression consequently exists as a very broad category defined by somewhat arbitrary criteria, as such the validity of such categories to

capture the complexity of the spectrum of mental health conditions has been questioned (Kinderman and Cooke, 2017).

Genome-wide association analysis provides support for understanding depression as a continuum of symptoms rather than as a dichotomous disease construct. Wray et al., (2017) conducted a genome-wide association meta-analysis of 130,664 individuals with depression (based on a combination of clinical assessment and self-report measures) and 330,470 controls. Findings strongly implicate a biological component of depression risk and the complex role of multiple genetic elements. The 44 identified by Wray et al., (2017) loci have been implicated in a variety of other disorders and diseases, including significant correlations with all other psychiatric disorders. For example, depression and schizophrenia were shown to have shared underlying genetic associations. Wray et al., (2017) propose this evidence strongly counters diagnostic models of categorising psychiatric disorders. Additionally, Wray et al., (2017) observed that the genetic architecture of depression (in those clinically diagnosed) was very similar to that of current depressive symptoms within undiagnosed community samples. This adds further weight to the understanding of depression risk as a continuum; whereby individuals are exposed to varying levels of genetic risk in combination with non-genetic predisposing or protective factors leading to phenotypic expression somewhere along the continuum.

Depression exists along a spectrum of severity and episodes and symptoms can range from mild to severe and as such have varying impacts on relationships, stress and social, occupational and personal functioning. By definition, a diagnosis of depression necessitates clinically significant distress or impairment in social, occupational or other important areas of functioning for two or more weeks (APA, 2013). 'Subthreshold depression' (i.e. demonstrating some symptoms but not meeting diagnostic threshold) has also been the object of research. Angst and Merikangas (1997), demonstrated that over the course of a 15-year longitudinal study, nearly half of all participants that initially met criteria for depression but were no longer depressed at follow-up demonstrated subthreshold depression that tended to persist over time. As such, this group of individuals, although no longer demonstrating clinically significant levels of depression, was not entirely asymptomatic. Similarly, Merikangas, Zhang and Avenevoli et al., (2003) employing the same cohort, demonstrate that threshold and subthreshold symptoms (those not meeting duration or impairment criteria) are prevalent, recurrent and distressing. Therefore, a further flaw of examining only those with

current diagnoses is that individuals with subthreshold or residual symptoms are overlooked within research.

While keeping in mind the criticism of the diagnostic approach, for these purposes, the symptoms and criteria specified within the DSM-5 (APA, 2013, see Figure 1.1) will be referred to in order to align with existing research. However, in order to avoid categorical diagnoses, the following empirical research studies employ the Mood and Feelings Questionnaire (Angold and Costello, 1987). This self-report measure captures depressive symptoms, based on the criteria of DSM-III-R (APA, 1987), however, the measure is linear rather than categorical scale. This was perceived to strike a balance between the need to assess depressive symptomology while retaining spectrum rather than categorical classifications.

Identifying Wellbeing

Attempts to measure symptoms and other qualities associated with distress encapsulate part of an individual's experience. In order to ascertain a fuller picture, it is important to consider positive qualities too. If there is to be a move towards the consideration of mental health as a continuum, ranging from highly disabling disorder to positive states of wellbeing, it is important that research reflects this. However, a lack of distress is not necessarily equal to positive experience; conceptually wellbeing is not only the antithesis of depression. Although mental health is considered to be an integral component of wellbeing (WHO: MH ACTION PLAN 2013-2020), wellbeing is considered to be greater than the absence of mental illness (WHO, 1947). As described by Duckworth, Steen and Seligman (2005: 630) "…persons who carry even the weightiest psychological burdens care about much more in their lives than just the relief of their suffering. Troubled persons want more satisfaction, contentment and joy, not just less sadness and worry". Similarly, it is possible for individuals at the opposite end of the depression spectrum (little or no symptoms) to experience very limited wellbeing due to other factors.

Wellbeing is a culturally dependent construct and has been considered broadly to be a state allowing individual flourishing, quality of life and the satisfaction of needs and desires (WHO, 1997; Ryff, Singer and Love, 2004; Kinderman, Schwannauer, Pontin and Tai, 2010). Wellbeing is thought to encapsulate positive qualities and fitness rather than negative qualities and lack of fitness, as well as, covering multiple domains of life including psychological, physical, relationship and economic factors. Components of wellbeing are recognised

protective factors against disorders, for example: physical safety, security and positive environments (NHS Scotland, 2016). For example, meta-analysis indicated that positive wellbeing was related to reduced mortality in healthy populations and with reduced mortality in patients with renal failure and human immunodeficiency virus (Chida and Steptoe, 2008). Promoting wellbeing is important in order to maximise protective and minimise risk factors of disorders. This is of particular relevance to adolescence where the occurrence of mental illness increases sharply.

The overall Mental Health Action Plan (WHO, 2013) identifies the promotion of wellbeing to be of key importance when working towards the aim of preventing mental disorders. The WHO calls for the identification of risk and protective factors for mental health and wellbeing, in addition to the reconceptualization of mental health to include positive states of wellbeing in combination with indicators of illness. This is also important for the concept of recovery and living well (Schmolke, 2003).

The current Government's Mental Health Strategy (2017), reiterates the importance of promoting wellbeing, considering this to be a national priority and discusses early intervention, community and public involvement, as well as the need for integrating mental health promotion in educational settings. This again identifies early life as a crucial stage in the development of wellbeing, as such schools and local authorities are encouraged to develop a curriculum which promotes wellbeing and supports individuals who may be at particular risk. This is reflected in the development of educational policies in Scotland and the UK: Getting it Right for Every Child, established within The Children and Young People (Scotland) Act 2014 and Every Child Matters (2003).

In order to gain greater insight and a more holistic perspective, it is important, within health research, to consider positive measures of wellbeing and quality of life in combination with symptomology and distress. Traditional over-emphasis of pathology and distress neglects the importance of wellbeing factors and positive measures of quality of life. As such, this thesis will consider depression and anxiety symptoms as well as measures of wellbeing throughout.

Recruiting from Community Settings

A further consideration concerning capturing the breadth of the spectrum of experiences within research is recruitment strategy. By definition, recruiting participants into research from

clinical settings, such as those already in treatment or those on waiting lists will include only those individuals that have been identified as experiencing difficulties. The high prevalence however, of mental ill-health within the adolescent population (11%; Avenevoli et al., 2015), along with the breadth of the mental health spectrum, suggests that symptoms are likely present within individuals recruited from community settings. Furthermore, not all individuals are help seeking; Wang et al., (2005) found on average, an 11-year delay between symptom onset and contact with health professionals. The biggest predictor of treatment contact delay was age, earlier onset disorders were consistently related to contact delay; meaning that individuals who identified as experiencing difficulties at an earlier age were likely to have the longest delay prior to seeking treatment.

Lawrence, Johnson, Hafekost, de Haan, Sawyer, Ainley and Zubrick (2015) report nearly half (44%) of young people experiencing any mental disorder had not engaged in any kind of service. There are significant barriers to young people in terms of accessing treatment, Lawrence et al., (2015) report the most common reasons as being: not sure where to get help, could not afford it, preferred to handle by self or with family/friends, and could not get an appointment. A recent report by the charity organisation SAMH assesses the need and provision of mental health services within Scotland specifically for young people. Their report indicates significantly overstretched health service with 1,838 referrals to CAMHS services between January and March 2017 were rejected. Of particular relevance to the sample populations of this thesis is that NHS Lothian failed to meet the maximum 18-week waiting time target 48% of the time; and, 10% of patients in Lothian had been waiting over a year for CAMHS appointments (Gordon and Platt, 2017).

Therefore, sampling restricted to patient groups risks overlooking individuals who are not help seeking, those who may be demonstrating pre-clinical or sub-diagnostic symptoms, or those who exhibit resilience factors. Consequently, in the empirical studies described within this thesis, participants have been recruited from community rather than clinical settings in an attempt to capture a sample representing the full range of psychological symptoms of health and wellbeing.

1.3 Risk Factors

Neuroticism

Adolescence represents a sensitive neurodevelopmental window for the fostering of lifelong positive mental health (Marco, Macri and Laviola, 2011). Therefore, there is an urgent need to understand risk factors that are specific to the adolescent period. A major risk factor for anxiety and depression is neuroticism (Navrady et al., 2017). Neuroticism is one of the five personality dimensions in the Five-Factor Model of Personality and refers to a tendency to experience negative emotions and emotional instability, this has also been referred to as 'negative affectivity' (Clark, Watson and Mineka, 1994). Neuroticism is considered a largely heritable trait, sharing genetic factors underlying risk for internalising disorders (Hettema, et al., 2006). Empirical evidence supports neuroticism as an endophenotype for different psychiatric disorders. A meta-analysis examining the relationship between neuroticism and Axis I disorders found large effect size of the relationship (d=>.80, Cohen, 1988) between the two (Malouff et al., 2005). Furthermore, Khan et al., (2005) demonstrated, in a study of 7,588 adult twins large effect sizes between neuroticism, depression, general anxiety disorder and panic disorder.

Several models (vulnerability, common cause, spectrum, scar and state models, see Figure 1.3) have been considered to explain the relationship between neuroticism and psychological disorders. Ormel et al., (2013) conclude that, particularly in relation to anxiety and depression, common cause and vulnerability theories have the strongest empirical support. Evidence supports a robust association between neuroticism and both Axis I and II mental disorders including: depression, anxiety, somatoform disorders, schizophrenia, eating disorders; as well as, neurotic, borderline, avoidant, schizotypal, paranoid and antisocial personality disorders (see Lahey 2009). Kahn et al., (2005) demonstrated a large effect size for the association between neuroticism and anxiety disorders, supporting common cause theories of anxiety and depression.

The role of neuroticism as a potential causal factor in the onset of disorders has also been the subject of study. Research indicated that neuroticism and psychiatric disorders share the same genetic influences. Wray et al., (2017) genome-wide association analysis demonstrated the strong overlap of neuroticism and depression. Furthermore, the study demonstrated that the

genetic architecture of depression overlapped with all psychiatric disorders and that for example, depression and schizophrenia had a shared genetic basis.

Models	Necessary condition	Supportive evidence
Vulnerability	1) Prospective association, i.e. neuroticism predicts first-ever episode of any CMD after adjustment for baseline symptoms 2) Neuroticism interacts with environmental determinants (e.g., life stress) to produce CMD	1) Presence of a clearly explicated causal chain linking neuroticism to CMD onset 2) Neuroticism mediates effect of psychological treatment on reduction of CMD symptoms
Common cause	1) Common determinants (assessed concurrently or prior to personality) account for the association between neuroticism and CMD	Primary prevention of CMD does also reduce neuroticism, in excess of the state effect.
Spectrum	1) Strong and specific association between neuroticism and CMD 2) Neuroticism and CMD share determinants 3) Overlap in measurement content	1) Synchrony of change in neuroticism and CMD symptoms 2) Similar differential stability 3) Treatment is equally effective in reducing symptoms and neuroticism scores
Scar	Post-episode neuroticism higher than pre-episode neuroticism	
State	1) Cross-sectional association 2) Effective treatment of CMD reduces neuroticism 3) Synchrony of change in neuroticism and CMD severity	1) Prospective association

Figure 1.3: Theoretical models describing the relationship between neuroticism and common mental disorders (from Ormel et al., 2013)

Various studies have found neuroticism to predict the onset of first lifetime episodes of depression. For example, high premorbid neuroticism robustly predicts future onset of depression (Kendler et al., 1993 and 2004). Navrady et al., (2017) found that each one standard deviation increase in neuroticism was associated with an increase in risk of depression by an odds ratio of 3.61 while Kendler et al. (1993) predicted that 55% of the genetic risk for depression was shared with neuroticism. Hettema et al. (2006) furthermore found that neuroticism and internalizing disorders were highly genetically correlated in over 9,000 twins, suggesting that associations are largely due to shared genetic factors.

Similarly, Ormel, Oldehinkel and Vollebergh (2004) compared never depressed (mean neuroticism score of 16.96), history of depression (mean neuroticism score of 19.92), first episode of depression (mean neuroticism score of 20.66) and recurrent depressed (mean neuroticism score of 21.84) groups to show increasing neuroticism across the four groups. This study used data collected as part of a three-wave Dutch population-based study and indicates that neuroticism is highest in those with recurrent depression, is elevated during first episodes of depression but does reduce in those with remitted depression, although not dropping to the levels of those with no history of depression. Ormel et al., (2004) however, attribute this to be a consequence of residual symptoms rather than scar effect due to the premorbid time period being far greater than the post morbid period. A population-based Swedish study examined 20,692 adult same-sex twin pairs in a 25-year longitudinal study which demonstrated that neuroticism scores predicted major depression 25 years later, even after exclusion of those with prior depressive episodes (Kendler, Gatz, Gardner and Pederse,

2006). It is significant that, after controlling for age, sex and extroversion, each standard deviation increase in neuroticism was associated with a 31% greater risk for a first episode of major depression over the 25-year period (Kendler et al., 2006). Such evidence is a strong indicator of shared genetic factors whereby there is overlap between neuroticism and disorder or, neuroticism is a predisposing factor to disorder.

Longitudinal studies in adolescence have also implicated neuroticism in psychological illness. For example, a New Zealand birth cohort demonstrated the highest quartile of neuroticism scores in 15-21-year-olds were at a 225% greater risk for suicide attempts at age 14 than those in the lowest quartile even after controlling for depression and other related factors including stressful life events, socioeconomic status and other mental disorders (Fergusson, Woodward, and Horwood, 2000). The combination of high neuroticism scores and these other related factors was 60 times greater than neuroticism alone, suggesting that neuroticism is of particular relevance when considered in the context of additional risk factors (Fergusson et al., 2000). Data collected as part of the Scottish Bipolar Family Study identified that individuals who were at high familial risk for bipolar and depressive disorders demonstrated significantly higher neuroticism scores than a control group (Ganzola et al., 2017). Furthermore, this study demonstrates that those who developed depression over the follow-up period demonstrated higher premorbid neuroticism scores (median= 32.5) than those who remained well within this high-risk group (median=20.5; Ganzola et al., 2017). High neuroticism preceding depressive onset further implicates neuroticism as a mechanism of disorder development.

The relationship between neuroticism and depression has often implicated other factors. Kercher, Rapee and Schniering (2009) conducted a large-scale path analysis within a sample of adolescent girls, to demonstrate that neuroticism constituted a distal vulnerability for depressive symptoms 12 months later. In this model, experiencing negative life events and negative automatic thoughts fully mediated the effect of neuroticism on depression. Kuyken, Watkins, Holden and Cook's (2006) cross-sectional study likewise suggests that neuroticism is a risk factor for adolescent onset depression and this possibly increases brooding rumination as a mechanism of action in response to depressed mood. Genetic research aiming to disentangle issues of directional causality is ongoing and may improve understanding in the future.

There is discrepancy regarding the stability over time of neuroticism. Frequently, neuroticism (and personality more generally) has been considered a stable trait. However, a randomised

controlled trial of adult patients with moderate to severe depression examined neuroticism change in SSRI (paroxetine), CBT and placebo conditions. Those taking SSRIs demonstrated 6.8 times greater reduction of neuroticism compared to patients exposed to the placebo condition after controlling for improved depression symptoms (Tang, DeRubeis, Hollon et al., 2009). Patients exposed to placebo or CBT treatment demonstrated a reduction in depression symptoms but no change in neuroticism. Tang et al. (2009) also demonstrated that reduction of neuroticism during treatment predicted relapse rates in patients taking SSRIs. One explanation for this is that SSRIs elicited changes to neurobiological factors underlying neuroticism, which contributed to depression improvement, and the authors considered that personality change accounted for the advantage of SSRIs compared to the placebo. However, Renner, Pennix, Peeters, Cuijpers, and Huibers (2013) demonstrated that changes in depressive state likely instigated changes in neuroticism, rather than the other way around, and that neuroticism itself was not directly modified by treatment.

It has been argued that the utility of measuring neuroticism is limited due to overlap of the construct with depressive symptoms. Consequently, Bowen, Baetz, Leuschen and Kalaychuck (2011) divide neuroticism scales into components that measure mood instability and traits of negative affect; they consider the latter to be confounded with disorder symptoms (i.e. depression/anxiety). However, the dissociation of depression and neuroticism change following SSRI versus placebo treatment demonstrated in Tang et al's (2009) RCT indicates that the relationship between depression and neuroticism cannot be explained by conceptual overlap.

Despite some lack of clarity in the mechanisms underlying neuroticism, it remains a construct of importance, particularly in less-researched populations, due to the extent of association of neuroticism with various mental and physical health conditions. Neuroticism has been considered a means of indexing risk and a general risk factor influencing the onset and course of psychological disorder (Klein et al., 2011). Lahey (2009) provides a discussion regarding the powerful predictive value of neuroticism in relation to longevity, psychiatric and physical health disorders. Consequently, within this thesis, neuroticism will be considered as a heritable factor which may index risk for depression and anxiety. The overarching aim of this body of work is to consider mechanisms contributing to psychological health and wellbeing above and beyond neuroticism.

Cognitive Vulnerability

Beck's (1967) cognitive theory proposed that emotional disorders and vulnerability to emotional disorders is characterised by dysfunctional schemas (internally held representations of experiences, stimuli and concepts) revolving around depressive themes, such as loss and failure. Schema are considered to be activated by internal or external events which influence encoding, storage and retrieval aspects of information processing (Beck, 1967). In relation to depression, this theory posits that depressed individuals exhibit a cognitive pattern involving a negative view of the self, the world and the future (the negative triad), arising from dysfunctional schema. Beck's (2008) theory posits that the activation of negative schemas constitutes cognitive vulnerability and predicts information processing biases which contribute to the development, maintenance and recurrence of depression. If such a vulnerability exists and results in biased information processing, such cognitive biases must be demonstrable. Empirical evidence supports this and links cognitive biases with depression in adults in a variety of realms of information processing including attention, interpretations and memory (Williams, Watts, MacLeod and Matthews, 1997).

Everaert, Duyck and Koster (2014) conducted path analysis (with a young adult sample; mean age=20.3 years) the results of which provide support for a combined cognitive bias model, whereby cognitive biases act in an integrative manner to impact functioning. Depression was significantly associated with attentional bias (γ=.25 (SE=.12), $p<.05$,) and interpretation bias (γ=.61 (SE=.09), $p<.001$). While attentional bias and interpretation bias were associated with memory bias (β=.24 (SE=.10), $p<.05$; and, β=.49 (SE=.10), $p<.001$, respectively). This evidence indicates that, during depressed states, attentional bias orients attention towards negative information which is followed by more negative interpretations of this information which in turn results in enhanced recall of negative material (Everaert et al., 2014). Consequently, despite acting within differing modalities, this research indicates an interplay between attentional biases and interpretation biases in the regulation of information recall.

A recent review identified an association between depression and attentional bias within depressed youth; however, it warns that effects may be better explained by anxiety (Platt et al., 2017). Platt et al., (2017) calls for greater investigation of memory bias within adolescence due to inconsistencies of findings thus far. Furthermore, interpretation bias has demonstrated initial evidence to indicate it may play a causal role in the onset of depression, although published studies are limited in number (Platt et al., 2017 reviewed only three such studies).

As such, biases of memory and interpretation will be the focus of this thesis. Consequently, when exploring cognitive biases, this thesis will focus on examining biases in the realms of memory and interpretation.

Biophysiological Risk Factors

Holistic approaches to care identify the importance of physical health in the context of mental health and wellbeing. Increasingly, the relationship between physical and mental health is being recognised within research and healthcare. This may be due to the identification of significant health inequality experienced by individuals with psychiatric disorders, who demonstrate a relative risk of mortality of 2.22, encapsulating a median of 10 years reduced life span (Walker, McGee and Druss, 2015). Although the literature tends to focus on the severe mental illnesses such as psychosis and schizophrenia, Walker et al., (2015) demonstrate that all-cause mortality (natural and unnatural) was significantly higher in those experiencing mood disorders. They suggest that those with mental disorders may not be experiencing the increased life expectancy of the general population. Furthermore, while psychosis demonstrated significantly higher relative risk of mortality compared to depression and anxiety, the overall prevalence of depression and anxiety accounted for the greatest contribution to deaths overall. Importantly this identifies that it is not only severe and enduring mental health conditions, that demonstrate significant disparity in mortality but that the most prevalent mental illnesses, anxiety and depression, are also of significance here.

HPA-Axis Regulation

One area of interest, that has been implicated as a potential mediating factor between psychiatric and physical disease, is the stress response system. Stress has been established as a precursor and marker of illness and disease (McEwen and Stellar, 1993). The HPA-axis comprises the neuroendoctrine pathway of the stress-response system (see Figure 1.4). The glucocorticoid hormone, cortisol, is a primary product of the HPA axis, the production of which supresses the HPA-axis (Lee, Kim and Choi, 2015). Cortisol is produced in response to stress as part of the evolutionarily driven stress-response system.

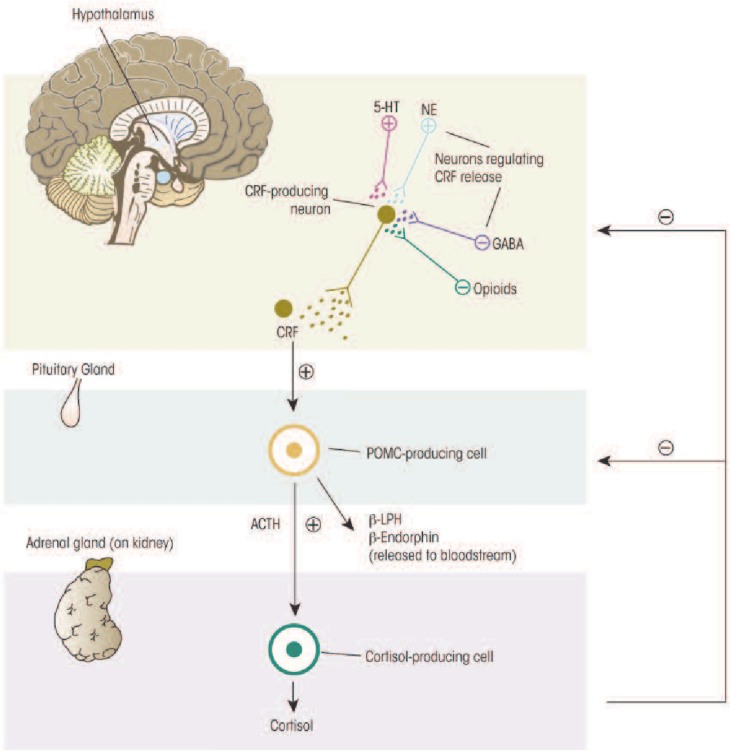


Figure 1.4: Major components of the stress response mediated by the hypothalamic-adrenal (HPA) axis; Stephens and Wand (2012)

Measuring Cortisol

Due to natural variations of cortisol levels, a consequence of the circadian rhythm and environmental stressors, which have the potential to elevate cortisol levels up to one hour after onset prior to returning to baseline, concentrations of cortisol in saliva can only assess current concentrations of cortisol and are vulnerable to environmental changes. Typically, cortisol follows a circadian rhythm whereby concentrations are high in the morning, peak shortly after wakening (known as 'cortisol awakening response' or CAR, see Figure 6) then decrease slowly throughout the day and are at their lowest in the evening (see Chung, Son and Kim, 2011 for detailed explanation of molecular regulation and Figure 6). Consequently, previous research has examined cortisol at various time points and the magnitude of change between time points.

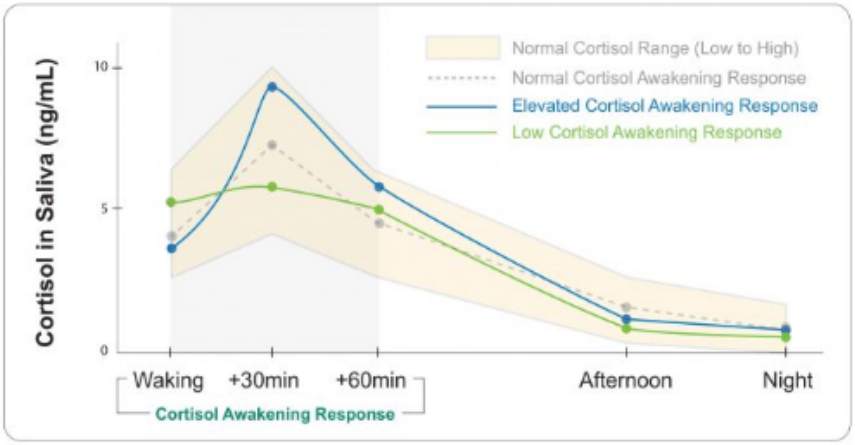


Figure 1.5: Cortisol Diurnal Profile (https://practitioner.researchnutrition.com.au/functional-tests/dutch-plus/)

Analysis of cortisol levels within hair samples has been employed to measure more general exposure to cortisol over time (see Figure 1.5). For example, cortisol levels in neonates' hair was related to the amount of time spent in neonatal intensive care (Yamada et al., 2007) and higher hair cortisol levels were found in a chronic pain group compared to healthy controls (Van Uum, Suave, Fraser, Morley-Forster, Paul and Koren, 2008). Karlen et al (2011) demonstrated that levels of cortisol were related to experience of serious life events, perceived stress and psychological problems during three months prior to assessment in university students and conclude that cortisol measured in hair can be a useful retrospective biomarker of increased cortisol production reflecting exposure to major life stressors and psychological illness. Consequentially, cortisol in hair may serve as a valid longitudinal biomarker of chronic stress.

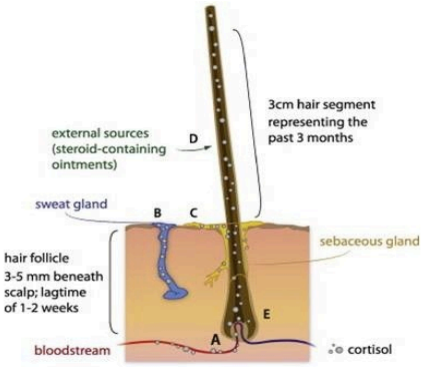


Figure 1.6: Hair exposure to cortisol; Lee, Kim and Choi, 2015

Cortisol and Health

This stress response through the HPA axis is proposed a be a major physiological mechanism through which stress influences disease risk (Karlen et al 2011; Cohen, Janicki-Deverts,

Miller, 2007; Cacioppo, Bernston, Malarkey, Kiecolt-Glaser, Sheridan, Poehlmann, Burleson, Ernst, Hawkely and Glasser, 1998). A variety of cortisol measurements have been examined in relation to health outcomes, which have implicated cortisol in a wide variety of mental and physical health outcomes (Chrousos and Gold, 1992; Hoyt, Craske, Mineka and Adam, 2015). Meta-analysis by Adam et al., (2017) has demonstrated that flatter diurnal cortisol slopes are associated with poorer health outcomes (see Figure 5). Consequently, the regulation of cortisol has been proposed as a mechanism mediating the relationship between stress and development of psychological and physical health disorders.

Research particularly indicates a relationship between mood and cortisol; specifically, negative affect has been associated with dysregulation of daily cortisol secretions as measured via HPA axis activity. Although, the strength of association is relatively small (r^2=0.01 in relation to depression and diurnal cortisol slope), waking cortisol was associated with a significantly greater likelihood of having a current internalising disorder (r^2=0.005, p=0.002). Both current and past internalising disorders were reliably associated with flatter slopes. However, this analysis was conducted on a relatively small sample (N=182), 19 of whom experienced current internalising disorders (Adam et al., 2017). This meta-analysis also demonstrated non-significant relationships between three measures of cortisol (waking cortisol, cortisol awakening response and cortisol slope) and personality trait neuroticism.

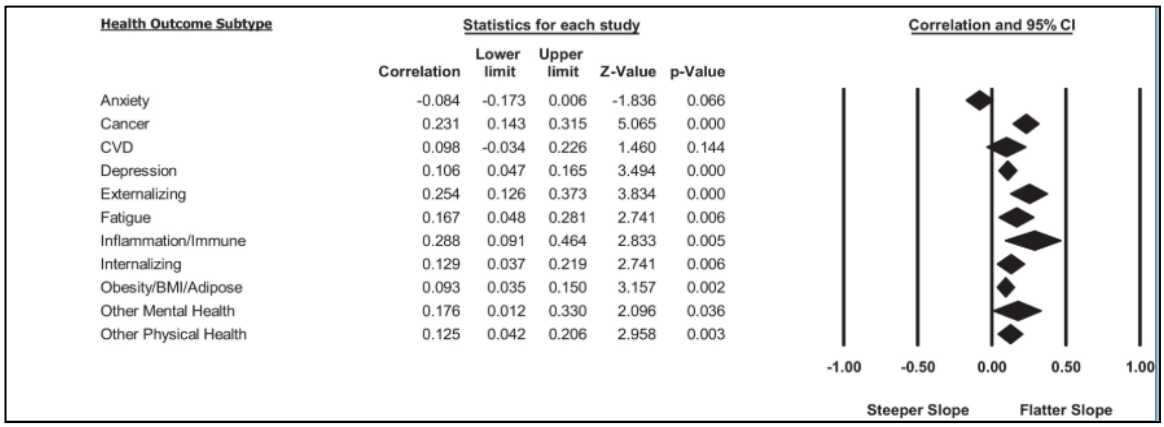


Figure 1.7: Adam et al., (2017) Forest Plot of Associations Between Individual Health Outcomes and Diurnal Cortisol Slopes (Pearson r).

Hebert, (2013), calls for the "serious consideration" of the role of cortisol in relation to depression. This review indicates that cortisol contributes to genetic variants for the risk of depression and implicates cortisol as a mechanism by which environmental factors amplify risk. The influence of cortisol begins prenatally but continues throughout life and has a

significant impact on psychological and biological variables (Herbert, 2013). Depression may be considered a consequence of sustained hyperactivity of the stress-system due to CRH overdrive, hyperactivity of the adrenergic system and/or excessive circulating glucocorticoid hormones. Chronic hypoactivation of the stress-system may lead to inertia, excessive fatigue and amotivational syndromes (Herbert, 2013).

Dysregulated cortisol rhythms and elevated morning and evening cortisol have consistently been reported as a risk factor, or consequence of MDD. Due to the recent emergence of this field of study and a scarcity of studies that have addressed cortisol in adolescent populations, the findings are not robust and often lack replication. Goodyer, Herbert, Tamplin and Altham (2000) examined a sample of 180 adolescents at high risk due to stressful life events, demonstrated that high peak (greater than the 80th percentile of the group mean) cortisol levels (all measured at 8am) were predictive of onset of depression (odds ratio=7.1, p=0.008, 95% CI: 1.67-30l.4). One limitation of this research is that due to the variation in individuals' waking preference and circadian rhythm, there is likely to be variability in the timing of peak levels is likely between individuals. There is discrepancy within research of sampling times with some studies restricting sampling to specific times of day, whereas, other research has employed sampling at waking, allowing for variation in individuals' waking preferences. Furthermore, these findings may not be generalisable to adolescents who have not experienced stressful life events.

Findings within adolescent samples are inconsistent. For example, Goodyer et al., (2000)'s finding of a relationship between high peak cortisol levels in the morning with depression, is somewhat at odds with other research, which supports a relationship between flatter (reduced) diurnal cortisol production and depression (e.g. Owens et al., 2014). However, Ulrike, Reinhold and Dirk (2013) found that in a sample of 131 female adolescents (mean age=15 years), higher cortisol awakening response was found in those who were currently depressed (n=63). As such, further examination of these factors is required. However, one possibility is that cortisol regulation comprises a significant risk factor for the onset of depression.

Distinction of the type of disruption of the HPA axis has been associated with(e.g. increased activity, decreased activity or dysregulated activity) has been demonstrated. A review by Tsigos and Chrousos (2002) discusses that a group of disorders have been characterised by hypoactivation of the stress system; this has been demonstrated in a variety of conditions including atypical depression, seasonal depression, chronic fatigue syndrome, obesity and

fibromyalgia. Conversely, hyperactivation of the stress response system has been related to a different spectrum of disorders, such as melancholic depression, anorexia nervosa, obsessive-compulsive disorder, childhood sexual abuse, obesity and severe chronic disease (Tsigos and Chrousos, 2002). As environmental factors (e.g childhood adversity and obesity) are considered risk factors for psychiatric disorders, pinpointing predictors of disorder onset is particularly complex requiring the disentanglement of the role of these factors and that of the stress response system. It is of particular importance to understand relation with HPA axis functioning in childhood and adolescence, where research is more limited and scarce, as development is considered to have a significant impact on the stress response system.

Sleep Duration and Disturbance

Sleep is largely driven by two processes. The circadian (~24 hour) rhythm, controlled by the superchiasmatic nuclei of the anterior hypothalamus regulates the production of hormones associated with sleep and arousal (e.g. cortisol and melatonin; Potter, Skene, Arendt, Cade, Grand and Hardie, 2016). The circadian system is considered to interact with a homeostatic sleep promoting system of accumulating sleep pressure during wakefulness (cueing sleep) and falling sleep pressure during sleep (see Potter et al., 2016 for detailed review, and Figure 1.8).

Adolescence is a period that is associated with a shift in sleep behaviour that cannot be accounted for solely by social factors; both the circadian and homeostatic processes have been implicated in this shift (see Hagenauer, Perryman, Lee and Carskadon (2009) for review). Cross-cultural research indicates that adolescents experience a sleep delay, whereby they demonstrate later sleeping preference and increased day time sleepiness (Hagenuaer et al., 2009, and Figure 1.9). Research from the Carskadon laboratory indicates that adolescents develop a delay in their circadian phase associated with developmental changes in circadian timing, alongside increased resistance to sleep pressure resulting in later bed times (Hagenauer et al., 2009).

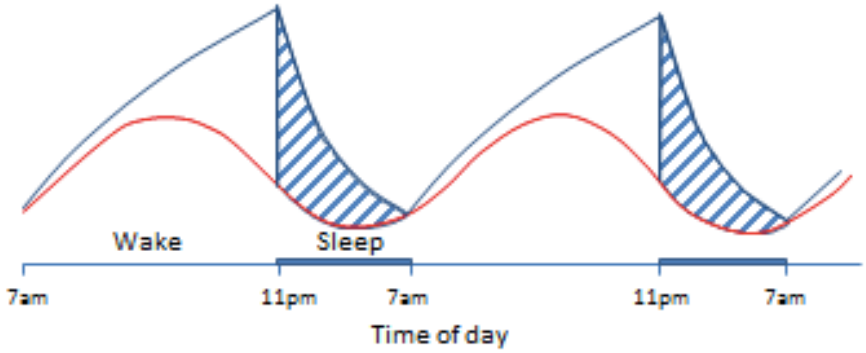


Figure1.8: Harvey and Peirson (2015), The Two Process Model of Sleep Regulation

Note: The process model of sleep: in this model when we sleep is driven by *sleep homeostasis* (the top line pictures in the picture above), or an increasing need for sleep, which grows the longer we have been awake. This is underwritten by our circadian processes (the red line) that control alertness and tiredness throughout the day. It is because of this system that we don't gradually get more and more sleep as the day goes on, but rather experience peaks and troughs in our tiredness. The shaded areas show average sleep time of a health adult.

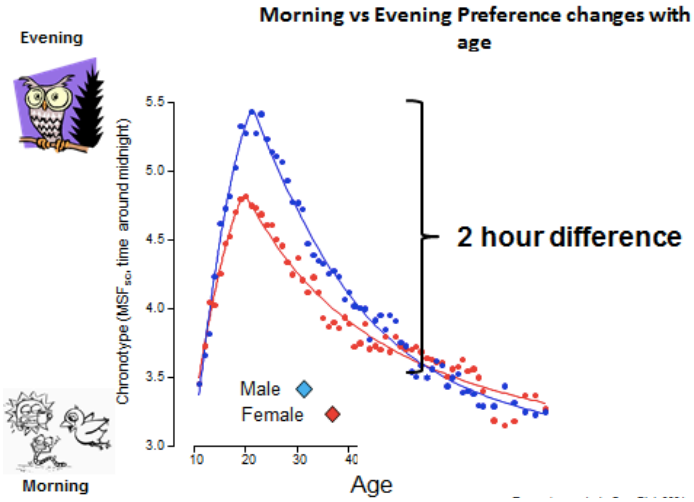


Figure 1.9: Harvey and Peirson (2015) Adolescent Chronotype Shift

Disturbed sleep is symptomatic of many of the psychiatric disorders, including depression and anxiety (APA, 2013). Insomnia (as well as hypersomnia) has been considered as more than a symptom or consequence of depression but a factor implicated in the onset and maintenance of depression, as well as, attenuating treatment success and persistence into remission (Clarke and Harvey, 2013). Trials within adult patient groups have demonstrated higher efficiency when antidepressant treatment was combined with CBT for insomnia. However, applications of this to adolescent populations are scarce (Clarke and Harvey, 2013).

Evidence indicates that short sleep duration is harmful; this has been particularly demonstrated within adult samples. While evidence of this in adolescents is rare, Baum et al., (2014) demonstrated in a small sample of adolescents that sleep restriction (6.5 hours per night)

compared to healthy sleep duration (10 hours per night) experienced significantly higher levels of anxiety, anger, fatigue and confusion, alongside, reduced vigour. Differences in depression scores were not demonstrated. However, higher levels of irritability and problems with emotional regulation were demonstrated. As such, this study demonstrated that several nights of reduced sleep adversely affected mood and emotional regulation capacity. Although individuals did not demonstrate differences in depression, this may have emerged over a longer duration of time. Furthermore, depression within adolescents has been considered to manifest as irritability, and problems of emotional regulation have also been implicated in emotional disorders (APA, 2000; Baum et al., 2014). Baum et al., (2014) have demonstrated a causal relationship between reduced sleep and psychological functioning.

Additionally, Fredriksen, Rhodes, Reddy and Way (2004) employed a longitudinal design that showed that reduced sleep significantly predicted an increase in depression symptoms and reduced self-esteem and academic performance. This study however, measured sleep as a one item self-report measure, which the authors note 'was not ideal'. Such measurement limits the understanding of nuanced details of sleep and does not consider quality of sleep.

Whitmore and Smith (2018) found that adolescent 9th-12th graders (approximately aged 14-18), achieving five hours or less sleep per night were 81% more likely to report suicidal ideation compared to those achieving 8 or more hours sleep (AOR=1.81 [1.62 and 2.02]). This relationship was maintained when controlling for demographic and behavioural factors, including depression. This study demonstrated a dose effect, students sleeping 6-7 hours per night were 17% more likely to experience suicidal ideation (AOR=1.17 [1.06, 1.29]). Of note is that, after stratifying the data based on depressed or non-depressed status of participants, only slightly improved predictions of suicidal ideation were observed. This indicates that sleep is a factor of key importance for the wellbeing of adolescents across the spectrum of mental health.

Research indicates a bidirectional relationship between mood and sleep. Associations between sleep disturbance and mood disorders are well established in adulthood; fewer studies however, examine this relationship in adolescence. A recent review indicated inconsistencies of findings in relation to adolescents, particularly relating to subjective versus objective measurement of sleep behaviour (Gregory and Sadeh, 2012). Typically, adolescence is associated with a change in sleeping patterns where by individuals tend to develop a preference for later sleep and waking times (Urrila, Puanio, Palomaki and Marttunen, 2015; discussed in

more detail in Chapter Five). This shift in combination with the increase of prevalence of mood disorders means it is of key importance that adolescents are studied separately from children and adults.

1.4 Thesis Overview

The primary focus of this thesis is adolescent depression. However, due to its conceptual relation to, and high comorbidity with anxiety, anxiety will also be measured and discussed. Furthermore, due to the importance assigned to wellbeing and the promotion of wellbeing in order to enhance quality of life and protect against illness, wellbeing will also be measured as one of the key outcome variables. In order to capture the continuum of these components, linear rather than categorical measures will be employed during each empirical study. Neuroticism is a personality trait that has been robustly associated with affective disorders (depression and anxiety) and has been considered to be a vulnerability factor. The overarching aim is to identify factors that may constitute markers of disorder, or those which may provide opportunity for future development of prevention or early intervention treatments.

In order to do this, contributions of factors that have been identified by previous research as potential vulnerability factors or markers of depression, but that lack robust and consistent evidence in the adolescent population will be addressed. Cognitive biases, sleep behaviour and the stress-response system will be examined in turn in relation to their relationship to mental health symptoms of depression, anxiety and wellbeing. The personality trait neuroticism will be considered throughout in terms of its relationship with both symptoms and the examined factors. The involvement of attachment (Chapter Three), negative cognitive style (Chapter Three and Four), stressful life events (Chapters Four and Five) and emotional regulation (Chapters Four and Five) will also be discussed.

Following this general introduction, Chapters Two to Five describe empirical research studies conducted with adolescent participants recruited from Scottish Secondary Schools. I recruited participants and completed the data collection and analysis described throughout this thesis (with the exception of the lab analysis of saliva and hair samples; see details in Chapter Four). Analyses are based on data collected from two distinct samples. Chapters Two and Three relate to the first sample, while Chapters Four and Five relate to the second sample. As far as I am aware there is no overlap of individuals within the two samples, although some participants were recruited from the same schools in both samples. Chapter Two and Three describe an empirical study conducted in order to assess contributions of the cognitive biases of memory

and interpretation to symptoms of depression, anxiety and wellbeing. Chapter Four describes an empirical study examining cortisol production and stressful life events and their relationship with depression, anxiety, and wellbeing, as well as neuroticism and emotional regulation. Chapter Five will report an empirical study that examines components of sleep in relation to depression, anxiety and wellbeing, as well as neuroticism and emotional regulation. Chapters two and three are based on the same sample while four and five are based on Chapter Six will comprise a general discussion of the thesis as a whole as well as its implications, limitations and conclusion.

Chapter Two – Investigating the predictive contribution of cognitive biases, in relation to neuroticism, to symptoms of depression, anxiety and wellbeing

A paper based on this study has been published: Smith, E.M., Orchard, F., Reynolds, S., Whalley H.C., and Chan, S.W.Y., (2018), Cognitive biases predict symptoms of depression, anxiety and wellbeing above and beyond neuroticism in adolescence, *Journal of Affective Disorders*, 241:446-453, DOI: https://doi.org/10.1016/j.jad.2018.08.051

2.1 Introduction

The previous introductory chapter introduced the concepts that will be examined within this chapter, namely, the proposition of cognitive bias comprising a vulnerability to depression and anxiety. As discussed in Chapter One, depression represents a significant global problem, which frequently originates during adolescents (WHO, 2017; MQ, 2016). Importantly, earlier onset of depression predicts longer episodes, a more severe and enduring course, poorer recovery and higher recurrence rates (Dunn and Goodyer, 2006). Including measures of positive aspects of psychological health is particularly important in adolescents as adolescence represents a sensitive neurodevelopmental window, with the opportunity to foster lifelong positive mental health (Marco, Macri and Laviola, 2011). Consequently, there is a need to understand risk factors within the adolescent context. A major risk factor for anxiety and depression is neuroticism (Navrady et al., 2017). This Chapter will describe and discuss an empirical study examining the role of cognitive biases in explaining depression, anxiety and wellbeing in adolescents. In particular this study was interested to identify salient cognitive factors that could explain psychological difficulties and wellbeing above and beyond the known personality risk factor neuroticism.

2.2 Neuroticism

As discussed in Chapter One, familial risk and personality traits, such as neuroticism, are the main known risk factors for adult depression (Weissman, 2016; Navrady et al., 2017). Neuroticism has been considered a means of indexing risk and a general risk factor influencing the onset and course of psychological disorders (Klein et al., 2011). However, such heritable and biological contributions to depression are not universally considered to be directly amenable to intervention. Lahey (2009) provided a discussion regarding the powerful

predictive value of neuroticism in relation to longevity, psychiatric and physical health disorders.

Briefly, evidence supports a biological basis of neuroticism that is considered to be a largely heritable trait, sharing genetic factors underlying risk for internalising disorders (Lucinao et al., 2018, Hettema, et al., 2006). However, there is mixed evidence regarding the stability of neuroticism over time. Wray et al., (2007) examined genetic and environmental contributions to neuroticism across 22 years in over 20,000 individuals, to demonstrate that genetic correlations between measures over time were very high. Environmental contributions however, demonstrated lower stability over time (Wray et al., 2007). Additionally, neuroticism has been shown to reduce following antidepressant administration (Tang et al., 2009), suggesting that the impact of improving mood through treatment may also have a more general impact affecting neuroticism. Other interventions however, such as cognitive treatments, have not demonstrated changes in neuroticism (Tang et al., 2009). As neuroticism is not universally considered modifiable, it is important to consider mechanisms that could explain the risk for psychological disorders beyond neuroticism that may be more amenable to intervention.

2.3 Cognitive Biases

Cognitive mechanisms are potentially more responsive and are targets of first-line treatments such as Cognitive Behavioural Therapy (CBT; NICE, 2005). However, existing cognitive treatments have demonstrated limited success in the adolescent population (Goodyer et al., 2017). CBT is based on adult models of depression and may be less efficacious for use in adolescence due to ongoing neurocognitive development and environmental factors in this unique developmental stage (Garber et al., 2016). Moreover, contributory factors may be age sensitive. As such, identifying cognitive features that predict depression in this age range, that may be amenable to modification, can potentially form a basis for improving or developing interventions with the potential for lifelong impact.

Negative cognitive biases of information processing have been proposed as mechanisms underlying the vulnerability, onset and maintenance of depression (Beck, 2008, Williams et al., 1997, Ingram, 1984, Joormann et al., 2007). Theoretically, mood-congruency bias predicts that information that is congruent to mood state, will be processed more efficiently, encoded more deeply and therefore, more easily retrieved (see Barry, Naus and Rehm, 2004). Consequently, within depressed mood, there is a predicted negative bias in relation to

information processing. Williams et al., (1997) proposed that negatively biased elaborative processing characterises depression. This necessitates increased allocation of cognitive resources to negative materials, resulting in the encoding of negative elaborations to memory, thereby enhancing memory for depression related materials. A vast quantity of research has examined cognitive biases in relation to depression in adults and identified the importance of assessing biases across multiple domains of processing. These have however been identified as a key limitation of research in the adolescent population (Everaert et al., 2015).

Autobiographical Memory Bias

Autobiographical memory is a component of memory related to personally experienced events and is heavily implicated in individuals' conceptualisation of the self (Williams, Barnhofer, Crane, Hermans, Raes, Watkins and Dalgleish, 2007). Such memories have been classified as specific (something that happened at a particular place and time and lasted for a day or less); or nonspecific (also referred to as overgeneral). Additionally, Williams and Dritschel, (1992) further classified non-specific memories into categories of categorical (pertaining to a class of memories, e.g. 'all the times I've failed exams') or extended (referring to an extended period of time e.g. 'the summer holidays'). Impairment in the recall of specific autobiographical memory has been robustly associated with adult depression (Williams et al., 1997).

When asked to recall a specific autobiographical memory, depressed individuals are more likely to recall an overgeneral memory. Sumner et al., (2011) implicated overgeneral memory in the onset of depression, demonstrating that overgeneral memory retrieval was predictive of depressive relapse in adolescents experiencing chronic interpersonal stress. However, studies of autobiographical memory and depression in adolescents, including clinical, community and at-risk samples, have produced mixed findings (Swales et al., 2010; Chan et al., 2007). Dalgleish et al., (2007) demonstrated that the relationship between depression and autobiographical memory recall was fully mediated by executive functioning; this was however, based on an adult eating disorder sample. Overall, there is a lack of consensus regarding the context and mechanisms of autobiographical memory performance and the majority of previous studies are based on adult patient samples.

Overgeneral memory has been implicated in various aspects of psychological functioning such as problem solving and imagining future events. Specific memory has been found to moderate the effect of negative mood disturbance on problem solving performance in previously suicidal

patients (Williams, Barnhofer, Crane and Beck, 2005). Problem solving abilities have been implicated in reduced social problem-solving skills related to interpersonal difficulties frequently experienced by individuals with depression (Goddard, Dritschel and Burton, 1996). Similarly, lack of specificity of recall has been associated with difficulty in imagining specific future events (e.g. Williams et al., 1996). Difficulties with future imaginations is of clinical significance due to hopelessness about the future being a key component of suicidal ideation and behaviour (O'Connor and Kirtley, 2018). Consequently, (in terms of both future imaginings and social problem solving) such bias may have implications for the onset and maintenance of depression.

Self-referential Memory Bias

A 'self-referent' memory bias has been robustly demonstrated, whereby information considered to be of personal relevance is more deeply encoded and more likely to be recalled than information that is not (see Symons and Johnson, 1997 for review). In depressed participants, depressogenic self-referent words and less positive words are considered to be more self-relevant and have been demonstrated to be more frequently recalled compared to anxious and healthy control participants. Tasks have been developed to assess this bias, known as Self-Referent Encoding Tasks. These tasks typically involve presenting individuals with positive and negative adjectives and asking them to judge whether or not they are self-descriptive. Subsequently, an incidental free recall task assesses recall (e.g. Derry and Kuiper, 1981; Hammen and Zupan, 1984). Evidence employing such tasks implicates a negative bias of self-referential information in depressed adults (e.g. Gotlib, Kasch, Traill, Joormann, Arnow and Johnson, 2004) and those considered to be high risk for depression (e.g. Alloy et al., 1997).

Studies examining such a bias in adolescence is less frequent, Timbremont et al., (2008) found no differences in memory biases between currently, never and previously depressed youth. In contrast, an early study of depressed youth employed a self-referent encoding task demonstrated memory bias in the recall (but not recognition) of negative compared to positive words (Zupan et al., 1987) Further, Cole and Jordan (1995) found that pupils with higher depressive symptoms recalled more negative self-referent words and fewer positive self-referential words and that this was related to depressive severity. A recent review (Platt et al., 2017) concluded that studies using self-referent encoding tasks have produced mixed findings and evidence of negative biases is inconclusive across clinical, at risk and community samples.

Therefore, further examination of this aspect of cognitive bias is warranted within the adolescent population. If individuals are more able to apply depressogenic content to their self-view, it is possible that this is a mechanism contributing to depression maintenance; this aligns with Beck's 'negative triad' conceptualisation regarding the negative view of the self, world and others (Beck, 1967, 1976). The lack of consistent findings and potential for intervention warrants further investigation of this phenomenon in adolescents.

Interpretation Bias

In addition to memory biases, negatively biased interpretation has also been associated with depression. Evidence indicates that depressed participants tend to interpret ambiguous information in a negatively biased way (e.g. Rude, Wenzlaff, Gibbs, Vane and Whitney, 2002). Orchard et al., (2016) demonstrated that adolescents with depression make significantly more negative interpretations of ambiguous social scenarios than non-depressed patient and community control groups. However, Micco et al., (2014) showed that interpretation bias modification reduced negative biases in depressed and control groups of adolescents, albeit with small sample sizes, but there was no associated change in anxiety or depression. Interpretation bias has also been identified within high-risk groups; Dearing and Gotlib (2009) found that daughters, aged 10-14, of depressed mothers responded more quickly to negative endings than control daughters in an ambiguous stories task. Although cognitive understanding of depression posits that negative interpretation bias is key in maintaining depression, and altering depressive interpretation is a component of cognitive behavioural therapy (Hollon et al. 2005), limited research has addressed this in the adolescent population and overall conclusions are limited by the scarcity of studies in the adolescent population.

Interpretation biases have not only been observed using ambiguous social scenarios, there has also been empirical evidence suggesting that deficits or biases in emotional processing of facial expressions are associated depression. Many studies of depressed adults demonstrate a negative bias in recognition of facial emotions (for review see: Kornreich and Philippot, 2006), fitting with Beck's conception of depression, and going some way to understanding interpersonal problems experienced by individuals with depression. Anderson et al. (2011) demonstrated that depressed individuals had reduced discrimination of anger, fear, sadness and happiness, compared to remitted and control participants. Interestingly, differences were not present in individuals taking anti-depressant medication, whose results reflected that of the

control group. Surguladze, Young, Senior, Brebion, Travis and Phillips (2004) demonstrated subtle differences in depressed participants' ability to recognise differing intensities of happy expressions. However, a distinctive pattern of impaired emotional recognition is not a robust finding, results are inconsistent between studies and once again, research among adolescents is scarce.

Findings of differences of facial emotional expression recognition within adolescent samples has been particularly inconsistent. For instance, Schepman et al., (2011) found no deficits of overall accuracy amongst depressed adolescents, although they were more likely to interpret low-intensity expressions as sad whereas non-depressed controls were likely to interpret low-intensity expressions as happy. Conversly, Joormann et al., (2010) demonstrated impaired accuracy of identification and that children at high familial risk for depression required greater emotional intensity of facial stimuli to accurately identify sad facial expressions. These inconsistent results could be due in part to varying methodology and ecological validity of tasks (Gotlib and Joormann, 2010).

Cognitive Style

Meta-analyses consistently demonstrate that rumination is predictive of the onset, severity and course of symptoms of depression (Mor and Winquist, 2002, Nolen-Hoeksema; 2000; and Nolen-Hoeksema et al., 1993). Negative attributional style is likewise considered to create a vulnerability following life events to depression (Ralph and Mineka; 1993). Rumination and dysfunctional attributional style are cognitive styles that have been associated with emotional disorders and cognitive biases. Rumination is considered to be a repetitive and passive self-reflective process with a focus on negative emotions whereby individuals struggle to shift to a new train of thought (Treynor, Gonzalez and Nolen-Hoeksema, 2003). A meta-analysis by Mor and Winquist (2002) found that the maladaptive process of rumination was the form of self-reflection most consistently related to depression, as opposed to adaptive self-reflective processes. Furthermore, rumination has been found to predict, onset, severity and course of depressive symptoms (Mor and Winquist, 2002; Nolen-Hoeksema, 2000). In adolescents, McLaughlin and Nolen-Hoeksema (2011) demonstrated that rumination fully mediated the association between anxiety and depression, identifying rumination as a transdiagnostic risk factor and target for intervention.

Similarly, attributional style may create a vulnerability or invulnerability following life events to depression. Depressive attributional style is considered to be the attribution of events as due to internal and stable causes (e.g. due to the self and being unchangeable); individuals with this attributional style are more prone to experience depressive reactions to negative life events than individuals who attribute events to external unstable causes (e.g. due to factors beyond the self and with the possibility for change; Seligman, Abramson, Semmel and von Baeyer, 1979).

Neuroticism and Cognitive Bias

Limited research has assessed neuroticism directly in relation to cognitive biases. However, Chan, Goodwin and Harmer (2007) demonstrated that never depressed young adults with high neuroticism exhibited an increased negative bias in relation to self-referential recall and facial expression recognition. Although there were no differences between high and low neuroticism groups in terms of recognition of descriptors, the high neuroticism group recalled fewer positive self-referent items. Conversely, Bradley, Mogg, Galbraith and Perrett (1993) found no significant effect of neuroticism on recall in response to a similar task. Furthermore, Chan et al. (2007) also demonstrated that a high neuroticism group required a higher intensity threshold of recognition of happy faces relative to the low neuroticism group only for happy facial expressions. Additionally, no relationship between neuroticism and autobiographical recall was demonstrated (Chan et al., 2007). Due to the limited availability of research directly examining neuroticism and cognitive biases, further examination of these components is warranted.

Aims of this Study

Some progress has been made in terms of understanding cognitive bias and how this relates to mood disorders in adolescents; only a few studies however, have been conducted and differing methodology makes reliable conclusions difficult to draw. Few studies have assessed multiple information-processing elements with the same sample. As such, further investigation is needed to increase understanding of this area, which is central to a fuller understanding of the impact of depression as well as its underlying mechanisms. The understanding of cognition serves as the basis of psychological therapy, such as cognitive behavioural therapy. Treatment in adolescents relies upon the adult model of CBT based on understanding of cognition within adults. As CBT has been demonstrated to be less successful in adolescence (TADS, 2014) it is possible that the underlying cognitive processes are also distinct. If cognitive bias is a driver

of mood disorders, a fuller understanding of cognitive processes and mechanisms are important in the development of effective treatments as well as ensuring that current treatments are effectively targeting the dysfunctional cognitive mechanism. Research in this area will also be beneficial in terms of prevention or early interventions, such as cognitive bias modification (see Hallion and Ruscio, 2011 for meta-analysis).

The overarching goal of this study was therefore to identify modifiable cognitive mechanisms that are predictive of adolescent mental health. This study aimed to recruit adolescents from community settings to examine whether cognitive biases can predict three outcome variables (depression, anxiety and wellbeing) above and beyond that of neuroticism. To overcome limitations of previous research, this study investigated, within a single sample, multiple cognitive factors including rumination, dysfunctional attitudes, and cognitive biases in self-referential and autobiographical memory, interpretation of ambiguous scenarios, and facial emotion recognition. It was hypothesised that measures of cognitive biases will predict depression, anxiety and wellbeing above that of neuroticism in adolescence, and that there may be distinct patterns of biases contributing to depression, anxiety and wellbeing.

This study therefore aimed to recruit adolescents from community settings to examine to what extent cognitive processes (attribution style, rumination and cognitive biases) are able to predict three outcome variables (depression, anxiety and wellbeing) within adolescents. Secondly, to consider the contribution of cognitive processes in relation to neuroticism. This study has examined contributions of cognitive bias factors independently from neuroticism in order to distinguish the influence of cognitive factors distinct from underlying personality risk. We hypothesised that cognitive measures will predict depression, anxiety and wellbeing above that of neuroticism in adolescence, and that there may be distinct contributions of biases to depression, anxiety and wellbeing.

2.4 Methods

Participants

This study recruited a total sample of 151 adolescents aged 12-18 years (mean=14.9, S.D. = 1.52) from Scottish secondary schools (58.6% female and 70.3% White British). See Table 2.1 for demographic details.

Table 2.1: Participant Demographics

Measure	Response	Percentage
Gender	Male	41.4
	Female	58.6
Ethnicity	White British	70.7
	White Other	12.1
	Asian British	9.1
	Asian Other	5.1
	Black British	2.0
	Black Other	1.0

Due to some participants failing to complete the online psychological measures and school time constraints for completion of face-to-face tasks, the sample size varies between measures. Based on the Mood and Feelings Questionnaire cut-off points for depression (>29; Daviss et al., 2006), 15 participants (15.15%) reported symptoms of depression that reached clinically significant levels. Those scoring above cut-off did not significantly differ from those below cut-off in terms of age ($t(97)=0.389$, $p=.691$), or gender ($t(97)=1.256$, $p=.212$).

Sample Size

A power calculation determined that a sample of 98 for up to 6 predictors in a regression model to detect a medium effect size ($f^2=0.15$, α error probability=0.05, 1-β error probability=0.80). Table 2.2 shows the sample size for each study element.

Table 2.2: Sample size for each study component

Measures	Number of Participants
Demographic Information*	151
Psychological Measures**	151
Ambiguous Scenarios Task	103
Self-Referential Recall Task	102
Autobiographical Memory Task	102
Facial Recognition Task	101
All Tasks and Measures	99
*refers to age, gender, ethnicity; **refers to MFQ, SCAS, BBC, EPQ-N, RRS and DAS	

Measures

All of the measures below are standardised measures that have been used in previous research with adolescents and have been considered valid and reliable. Their reliability in this sample was checked and confirmed with high Cronbach's alphas, as reported below.

<u>Mood and Feelings Questionnaire-Child Version (MFQ; Angold and Costello, 1987)</u>

This 33-item self-report scale measures mood symptomology as assessed by a 3-point scale. The MFQ measures severity of depressive symptomology as a unidimensional construct based on diagnostic criteria of the DSM and ICD diagnostic systems (Sharp, Goodyer and Croudace, 2006; Angold and Costello 1987). Internal consistency in the current data set was high, with Chronbach's α=.94. Although not employed within this study, previous research has demonstrated that a score of 29 distinguished individuals with diagnoses of depression (Daviss, Birmaher, Melhem, Axelson, Michals and Brent, 2006). Employing this criterion, 18% of the current sample demonstrated clinically significant scores using this scale.

Spence Children's Anxiety Scale (SCAS; Spence, 1997)

The SCAS is a 44 item self-report questionnaire assessing six subscales of anxiety (social phobia, obsessive compulsive, panic/agoraphobia, separation anxiety, physical injury fears and generalised anxiety) Participants were asked to rate the degree to which they experience each symptom on a four-point scale. This questionnaire demonstrated high internal consistency within this sample, Cronbach's α=.92.

BBC Well-being Scale (Kinderman, Schwannauer, Pontin and Tai, 2011)

This 24-item questionnaire assesses general wellbeing over three key domains: psychological wellbeing, physical health and wellbeing, and relationships. This scale has been validated in a sample of 1,932 participants, of whom 228 were school-aged adolescents. In this study, this scale demonstrated excellent internal consistency (α=.92). Participants were asked to respond on a 4-point scale. Cronbach's α=.95, indicating high internal consistency within this sample.

Eysenck Personality Questionnaire-Neuroticism subscale (EPQ-N; Eysenck, Eysenck and Barrett, 1985)

Neuroticism was evaluated using the widely employed shortened form of the neuroticism scale (12 items), from the Eysenck Personality Questionnaire (Eysenck, Eysenck and Barrett, 1985). Internal consistency was found to be high, Cronbach's α=.85 on this sample.

Rumination-Ruminative Response Styles (RRS; Treynor, Gonzalez and Nolen-Hoeksema, 2003)

This ten-item scale captures rumination with subscales of reflection and brooding (five items each). This version of the ruminative response scale has removed 12 items from the original scale that were considered to overlap with items from the Beck Depression Inventory (Treynor et al., 2003). These items have been considered to capture rumination in terms of brooding

and reflection as well as coping whiles remaining neutrally valanced. Participants rated each item on a four-point scale from 1-(almost never) to 4-(almost always). In this sample, internal consistency was high with Chronbach's α=.89.

Dysfunctional Attitudes- Dysfunctional Attitudes Scale: 24 Item Version (DAS-24; Power, Katz, McGuffin, Duggan, Lam and Beck, 1994)

Power et al. (1994) shortened the original DAS (Weissman and Beck, 1978) to 24 items. This version has three subscales, representing factors of vulnerability: achievement, dependency, and self-control. Internal consistency in this sample was high, with Chronbach's α=.85.

Self-Reference Recall Task (Kelvin et al. 1999)

This task assessed biases of self-referential memory recall and consisted of 12 positive (e.g. 'successful') and 12 negative (e.g. 'pathetic') adjectives. P This task has been previously employed in research literature and significant differences in this task were demonstrated Participants indicated the extent to which each word described them on a four-point scale (1= 'not at all like me', 4= 'very much like me'). To counter primacy and recency effects, three neutral adjectives were included at the beginning and end, responses to which were excluded from analysis. Ratings were subsequently recoded as either: not self-referent ('not me'), or self-referent ('me'). Participants were subsequently asked to recall descriptors in a free recall task. Participants had not initially been aware of the memory component of the task in order to increase the ecological validity of the task. A proportional score reflecting overall positive bias to the task in both 'me' and 'not me' conditions was calculated (Connolly, Abramson and Alloy, 2016). This was calculated by subtracting the number of correctly recalled negative words from the number of correctly recalled positive words and dividing this by the total number of correctly recalled words. Therefore, positive scores reflect a positive bias whereas negative scores indicate a negative bias.

Autobiographical Memory Test (AMT; Williams and Broadbent, 1986)

This task was used to assess autobiographical memory recall and has been extensively used within research and has previously demonstrated good psychometric properties (Griffith, Kleim, Sumner and Ehlers, 2012). Participants were presented visually and orally with five positive (relaxed, lucky, excited, relieved, loved) and five negative (hopeless, failure, sad, angry, lonely) cue words, in a randomised order, and asked to recall a specific memory. Specific memory was defined as 'a memory of a particular event that occurred on a particular day which could be from a long time ago or very recently and could be something very

important or something very ordinary'. Participants were given 60 seconds to produce a memory, which they then verbally described under no time condition. This task was audio recorded and later coded for level of specificity. Participants were given two practice trials, where they were given feedback and prompted to recall a specific event if their memory did not fit the criteria of a specific memory. Practice items were excluded from analysis. Responses to positive and negative cues were recorded as either specific (e.g. 'My friend took me to the Manchester derby on my birthday.') or overgeneral (e.g. 'During the summer holidays when there was no one around.') based on whether they meet the definition of specific memory stated in the instruction. The total number of specific and overgeneral memories was calculated.

Ambiguous Scenarios Task for Depression in Adolescents: (AST; Orchard, Pass and Reynolds; 2016)

Participants were presented with 20 hypothetical ambiguous scenarios and asked to consider each as happening to them and imagine the outcome. For example: 'You see a man running down the street and think about why he is running'. Participants were asked to (i) write down their imagined outcome, and (ii) rate its pleasantness on a 9-point scale (1= 'extremely unpleasant' vs 9='extremely pleasant'). No time limit was given for this task. In this study, only the written descriptions were included in analysis due to the relatively lower internal reliability of the rating scale in this sample (α=.68). Written descriptions were coded as positive, negative, neutral or mixed. In line with previous research (Orchard et al., 2016), only positive and negative interpretations were included in analysis. Overall bias scores were calculated by subtracting the number of negative responses from the number of positive responses. Therefore, positive scores reflect a positive bias whereas negative scores indicate a negative bias.

Facial Expression Recognition Task (Chan et al., personal communication)

This computer task, adapted by Chan et al. (personal communication), presented five emotional expressions: anger, disgust, fear, happy and sad. Stimuli were morphed using Morpheus Photo Morpher v3.17 software, from 0-100% intensity at incremental increases of 10%, with 0% reflecting a neutral expression and 100% reflecting the full intensity of expression. Stimuli depicted facial expressions of male and female, White and Asian faces. In total 220 stimuli were presented in a random order across five blocks. Each face was presented for 500ms against a black background preceded by a fixation cross of 100ms. Participants were asked to identify the emotion displayed by a key press with no time restriction. Prior to

the experimental trials participants completed six practice trials, which were excluded from analyses. Mean accuracy of identification was computed across each emotion.

2.5 Ethical Considerations

This study obtained ethical approval from the University Research Ethics Committee and local educational authorities (see Appendix). Written permission was obtained from principal teachers of participating schools and a liaising teacher was identified. A briefing session was arranged for school pupils that included distribution of Information and Parental Information Sheets. In most cases participating schools wrote to or emailed parents to inform them that this research study was taking place prior to the briefing session. Written consent was obtained from participants (and their parents if they were under 16; see Appendix). Participants, parents and teachers, had opportunities to ask questions throughout and were provided with contact details for the researcher if they had any questions or concerns at a later date.

This ethics procedure differed for one participating school. Following discussion and approval from the University of Edinburgh's Ethics Committee, some participants were recruited from a boarding school. In these cases, for participants under the age of 16, written permission was obtained from the participants' house master/mistress who acted in loco parentis. The House Master/Mistress was the individual who took primary responsibility for individuals' welfare while at school and was therefore considered the most appropriate individual to provide consent on participants' behalf. In these cases (as above) pupils under the age of 16 also provided written assent and those over 16 provided individual consent.

This study respected the rights and prioritised the welfare of participants by ensuring that they understood the aims and methods of the study. Procedures of confidentiality, anonymity, secure storage of data and duration of data storage were explained to participants. In all cases consent and assent forms asserted the rights of participants to volunteer as a participant, to withdraw freely at any time without consequence, as well as, confirming that they had read and understood the information sheet and had been provided with opportunities to ask any questions.

In accordance with data protection guidelines set out by the University of Edinburgh, all electronic data stored in relation to this study has been anonymised. Participants remain identifiable only by participant number and any written record linking names to participant

number have been securely destroyed. Hard copies of data and consent forms are stored securely and separately.

At the end of data collection all participants were provided with a Debrief Sheet (see Appendix) which provided information regarding mental health support. Specifically, participants were advised that they may wish to speak to a trusted adult such as their parents, teachers or GP's if they had any concerns about their mood or mental health.

2.6 Procedure

Having received Ethical Approval from the University of Edinburgh, and subsequently Ethical Approval from Edinburgh Council, invitation letters were sent to the head teacher or principal teacher of all mainstream secondary schools within Edinburgh Council region (n=36). This correspondence outlined the overarching aims of the study, to improve understanding of adolescent mental health, as well as brief details of the nature of the study and the requirements of participation. Head teachers were encouraged to respond with any questions or if they were interested in supporting the study. Recruitment proceeded within each school that expressed interest (n=5). Unfortunately, one school withdrew following collection of online measures due to time constraints within the school, resulting in incomplete data collection from these participants and subsequent exclusion of their data from analysis.

Those interested pupils who requested consent forms were asked to return them completed on an agreed upon date. At this point they were provided with a participant number and a link to an online survey, hosted by Survey Monkey, to complete psychological measures (depression, anxiety, wellbeing, neuroticism, attachment, rumination, and dysfunctional attitudes) online within their own time. In addition, a suitable time for schools and participants was agreed for the researcher to return to conduct the face-to-face measures (assessing autobiographical and self-referential memory, interpretation of ambiguous scenarios and facial expression recognition), in all cases this was within one week of completion of online measures. Data collection during these sessions was conducted either one-to-one or in small groups (average group size ~5 participants). If participants had not completed the online survey by this point the researcher reminded them to complete this component. Due to the anonymisation of participants it was not possible to follow up with individual participants after this point in relation to any incomplete components.

2.7 Statistical Analysis

Missing data

Participants that had not completed any psychological measures were excluded from analysis (n=52), leaving a sample of 99 (see Table 2.2). Descriptive data relating to those participants who completed online psychological measures but did not complete cognitive tasks is presented below (see Table 2.3), this included the school that withdrew from participation due to time constraints. T-tests indicated no significant group differences between those who did not complete cognitive tasks in terms of age or psychological data, other than in relation to anxiety (SCAS). Those who did not complete cognitive tasks exhibited significantly higher anxiety scores than those who did ($t(143)=14.87$, $p<0.001$). Potential explanations for this are included within the discussion. In addition, three participants did not complete measures of facial expression recognition. Analyses were primarily conducted in IBM SPSS version 23 (SPSS Inc., USA). Due to a procedural error, item 13 from the BBC Well-being Scale was missing at random for the majority of participants (75%). Consequently, this item was excluded and the mean rather than total score was employed for further analysis. The reliability of this scale remained high ($\alpha=.95$).

Table 2.3: Descriptive Statistics of Participants with Missing Data

Variable	Mean	Standard Deviation
Age	15.11	1.49
MFQ	15.29	12.66
SCAS	64.59	13.02
BBC	65.57	14.42
Neuroticism	6.61	3.74
RRS	21.11	7.29
DAS	103.09	22.22
n=46, 78.26% female		

Checks of statistical assumptions

Tests of normality were conducted to verify the appropriateness of the chosen analysis which indicated assumptions of normality were met. Despite variables being highly correlated (see results), collinearity and tolerance statistics were within accepted limits (maximum VIF=1.50 and lowest tolerance =0.61; Field, 2009, p. 297).

False discovery rate (FDR) correction for multiple comparisons, calculated using R version 3.2.5, was applied to significance values of Pearson's correlations examining relationships between variables to control for familywise error rate (Benjamini and Hochberg, 1995).

Analysis comprised two stages. Initially, to identify salient predictors, backwards-elimination regressions were employed based on a criterion of F>=.100. Subsequently, surviving variables were entered in a hierarchical regression to examine their contribution in relation to neuroticism.

The impact of gender and age was assessed using t-test and Pearson's correlation, if tests demonstrated significant effects these variables were included in regression models.

Each regression model consisted of two steps to predict each dependent variable (depression, anxiety and wellbeing) in turn: 1. Neuroticism, 2. Cognitive variables surviving backwards removal regression. When age and gender were included within regression models, these did not impact any results and as such have been excluded as variables in order to preserve statistical power. Gender was included in models of anxiety as univariate tests indicated significant relationships between gender and anxiety.

2.8 Results

See Table 2.4 for descriptive statistics.

Table 2.4: Descriptive Statistics

Measure	Mean	Std. Deviation	Minimum	Maximum
Age	14.94	1.52	13	18
MFQ (Depression)	15.26	12.88	0	57
SCAS (Anxiety)	27.59	17.09	2	95
BBC (Wellbeing)	68.56	12.87	35	92
Neuroticism	6.24	3.41	0	12
RRS (Rumination)	21.11	6.41	10	40
DAS (Dysfunctional Attitudes Scale)	91.55	17.45	51	168
AMT: Autobiographical Memory Task (Specific)	7.36	1.95	2	10
AMT: Autobiographical Memory Task (Overgeneral)	2.04	1.66	0	7
AST: Ambiguous Scenarios Task Bias	1.33	5.76	-15	15
SRR: Self-Referential Recall 'Me'	0.33	0.29	-0.50	1
SRR: Self-Referential Recall 'Not Me'	-0.23	0.19	-0.56	0.50
Facial Expression Recognition: Anger	0.57	0.15	0.23	0.88
Facial Expression Recognition: Disgust	0.43	0.15	0.05	0.73
Facial Expression Recognition: Fear	0.60	0.12	0.25	0.80
Facial Expression Recognition: Happy	0.76	0.08	0.48	0.93
Facial Expression Recognition: Sad	0.67	0.20	0.15	0.95

Age and Gender Differences

Female participants reported higher levels of anxiety than male participants: $t(97)=2.83$, $p=.006$. No significant gender differences were demonstrated for depression ($t(97)=-1.22$, $p=.27$) or wellbeing ($t(96)=1.85$, $p=.07$). There was no significant correlation of age with depression ($r=.13$, $p=.20$), anxiety ($r=.06$, $p=.55$) or wellbeing ($r=-.13$, $p=.21$).

Identifying Salient Predictors

See Table 2.5 and Figure 2.1 for Pearson's correlation results, (age and gender controlled).

Pearson's correlations, controlling for age and gender, indicate a significant relationship between depression and anxiety and a negative relationship between depression and wellbeing. Neuroticism was positively correlated with depressive symptoms and anxiety, while negatively correlated with wellbeing. See Table 2 and Figure 1 for full results. Briefly, correlations demonstrated the expected pattern with higher depression and anxiety symptoms as well as lower wellbeing generally correlated with greater negative biases, reduced positive biases or more maladaptive processing. Mean accuracy bias scores of facial emotion recognition and autobiographical memory bias scores were not significantly correlated with any other variables.

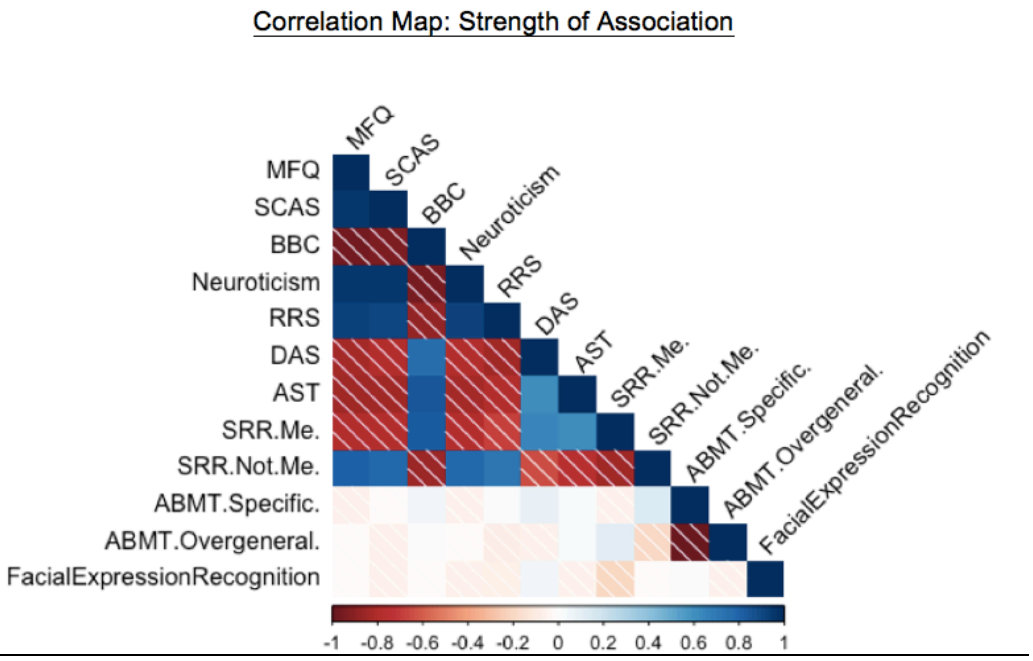


Figure 2.1 Strength of Association Heat Map

Note: Figure depicts Pearson correlation between variables. For simplicity, Facial Emotion Recognition Bias is represented by a mean across emotions. Diagonal stripes indicate negative correlation. MFQ refers to the Mood and Feelings Questionnaire, SCAS refers to Spence Child Anxiety Scale, BBC refers to the BBC Well-being Scale, DAS refers to the Dysfunctional Attitudes Scale, RRS refers to the Ruminative Response Scale, AST refers to the Ambiguous Scenarios Scale, SRR refers to the Self-Referential Recall task. ABMT refers to the Autobiographical Memory Test

Table 2.5:Pearson's Correlation Coefficient (r) between outcome variables and cognitive variables, controlling for age and gender

	Neuroticism (EPQ-N)	Rumination (RRS)	Dysfunctional Attitudes (DAS)	Ambiguous Scenarios (AST) Bias	Self-reference recall bias (SRR): Me	Self-reference recall bias (SRR): Not Me	Anger	Disgust	Fear	Happy	Sad	ABMT: Specific	ABMT: Overgeneral
MFQ	0.63**	0.58**	-0.51**	-0.41**	-0.35**	0.42**	0.21	0.01	0.14	-0.10	-0.07	-0.06	0.01
SCAS	0.71**	0.57**	-0.48**	-0.41**	-0.32*	0.34**	0.17	-0.08	0.08	-0.15	-0.15	0.00	-0.07
BBC	-0.57**	-0.48**	0.51**	0.48**	0.45**	-0.48**	-0.27	0.07	-0.02	0.16	0.03	0.04	-0.04

*Note: **indicates significance at p<0.001; *indicates significance at p>0.05. MFQ refers to the Mood and Feelings Questionnaire, SCAS refers to Spence Child Anxiety Scale, BBC refers to the BBC Well-being Scale*

Backwards-Elimination Regression

Variables surviving the regression model of depression were: rumination, dysfunctional attitudes and non-self-referential recall bias. Those surviving the regression model of anxiety were ambiguous scenarios bias and rumination. The ambiguous scenarios task bias, self-referential recall bias, dysfunctional attitudes and accuracy of anger identification were included in the wellbeing regression model (Supplementary Material).

Hierarchical Multiple Regression

See Table 2.6.

Depression

In predicting depression, neuroticism was a significant predictor (r^2 = .40 $p<0.001$, $\beta=0.35$). The addition of cognitive variables: rumination ($\beta=0.26$), dysfunctional attitudes ($\beta=-0.19$) and self-referential 'not me' recall scores ($\beta=0.17$), resulted in an increase of explained variance ($\Delta r^2=.12$, $p<0.005$; $r^2=.52$, $F(4,94)=25.49$, $p<0.001$). All included variables demonstrated significant beta values.

Anxiety

In the first block gender alone significantly predicted anxiety ($r^2=.08$, $p<0.05$), as did the addition of neuroticism (r^2 =.54, $p<0.001$, $\beta=0.55$). Inclusion of cognitive variables (ambiguous scenarios task bias ($\beta=-0.13$) and rumination $\beta=0.21$) demonstrated a small but significant increase of explained variance ($\Delta r^2=.05$ and $r^2=.59$; $F(3,95)=44.46$, $p<.001$,). Only neuroticism and rumination demonstrated significant beta values.

Wellbeing

In the first block of the model of wellbeing, neuroticism significantly predicted wellbeing (r^2 =.36, $\beta=-0.32$, $p<0.001$). Inclusion of cognitive variables: ambiguous scenarios task bias ($\beta=0.19$), self-referential recall 'me' bias ($\beta=0.18$), self-referential recall 'not-me' bias ($\beta=-0.16$), dysfunctional attitudes ($\beta=0.17$), and recognition of angry facial expressions ($\beta=-0.14$), demonstrated a significant r^2 change ($\Delta r^2=.19$, $p<0.001$; $r^2=.55$, $F(6,89)=17.74$, $p<0.001$). Only neuroticism, ambiguous scenarios task bias and self-referential recall 'me' bias demonstrated significant beta values

Table 2.6: Hierarchical Regression Models

Model		r	r^2	Adj r^2	F-value (df)	Unstandardised Coefficients B	SE B	β	t-value	Δr^2	ΔF (df)
MFQ		0.63	0.40	0.39	63.33(1,97) **					0.40	63.33
Step 1	Constant					0.45	2.12		0.21		(1,97)**
	Neuroticism					2.37	0.30	0.63	7.96**		
Step 2		0.72	0.52	0.50	25.49(4,94) **					0.12	8.18
	Constant					11.49	7.32		1.23		(3,94)**
	Neuroticism					1.32	0.35	0.35	3.82**		
	RRS					0.52	0.18	0.26	2.97*		
	DAS					-0.14	0.06	-0.19	-2.41*		
	SRR Not Me					11.68	5.25	0.17	2.23*		
SCAS		0.28	0.08	0.07	8.01(1,97)*					0.08	8.01 (1,97)*
Step 1	Constant					22.00	2.58		8.53		
	Gender					9.53	3.37	0.28	2.83*		
		0.73	0.54	0.53	55.93(2,96)**					0.46	96.01 (1,96)**
Step 2	Constant					4.44	2.56		1.73		
	Gender					0.76	2.56	0.02	0.30		
	Neuroticism					3.64	0.37	0.73	9.80**		
Step 3		0.77	0.59	0.57	33.33(4,94)**					0.05	5.49 (2,94)*
	Constant					-1.78	4.27		-0.42		
	Gender					1.83	2.47	0.05	0.74		
	Neuroticism					2.77	0.44	0.55	6.25**		
	RRS					0.55	0.22	0.21	2.54*		
	AST Bias					-0.39	0.22	-0.13	-1.82		
BBC		0.60	0.36	0.35	51.75(1,94)**					0.36	51.75 (1,94)**
Step 1	Constant					82.67	2.25		36.73**		
	Neuroticism					-2.27	0.32	-0.60	-7.19**		
Step 2		0.74	0.55	0.51	17.73(6,89)**					0.19	7.41
	Constant					65.94	7.96		8.58**		(5,89)**
	Neuroticism					-1.20	0.33	-0.32	-3.65**		
	DAS					0.13	0.06	0.17	2.10*		
	AST Bias					0.42	0.19	0.19	2.26*		
	SRR Me					7.82	3.74	0.18	2.09*		
	SRR Not Me					-10.98	5.91	-0.16	-1.86		
	Anger Acc					-12.05	6.68	-0.14	-1.80		

2.9 Discussion

Rumination, dysfunctional attitudes, and negative biases in ambiguous scenarios interpretation and self-referential memory, significantly predicted depression in adolescents above and beyond neuroticism. Neuroticism predicted around 40% of the variance of depression symptoms, 54% of anxiety symptoms and 35% of wellbeing, in line with previous research (e.g. Kotov et al., 2010 and Bartels et al., 2013). As hypothesised, adding cognitive variables significantly increased the explained variance of depression by 12%, anxiety by 5% and wellbeing by 19%. This supports previous findings that have associated negative cognitive biases with psychological outcomes. Orchard et al., (2016) demonstrated that biased interpretation was most negative for adolescents with a depression diagnosis and most positive for non-depressed controls. The current study highlights the salience of interpretation bias across psychological distress and wellbeing.

In an undergraduate student sample, Rude et al., (2002) demonstrated that including a measure of interpretation under a cognitive load condition increased predictions of depression scores by 11% for male participants and 0% for female participants. This increase is in line with the current findings; however, we did not identify such gender disparity. Rude et al. (2002) suggest that such divergence may be due to gender bias in self-report or that despite decreased negative biases in women, their subsequent behaviour or processing (such as rumination) increases their vulnerability to experience depression.

This study demonstrates, similar to findings of adult populations, that neuroticism is a key predictor of mental health outcomes (Lahey, 2009). Neuroticism was the strongest predictor of each mood variable; each standard deviation increase of MFQ was associated with a 0.55 increase of neuroticism. This magnitude is similar to findings of a meta-analysis of 33 population-based samples (Malouff, Thorsteinsson and Schutte, 2005). The predictive power of neuroticism cannot be overlooked as it indicates underlying biopsychological components of depressive disorders worthy of further neurobiological study to assess its expression and mechanism of action.

Like Zupan et al., (1987), this study indicates that greater depressive symptomology and anxiety was strongly correlated with a bias towards recalling negative self-referenced words. For non-self-referential words however, higher levels of depression and anxiety were related to recalling more positive adjectives. This suggests that symptoms of depression and anxiety are not associated with global negative biases, but a specific negative bias in relation to the self. This is consistent with findings of more negative self-perceptions in young people at risk

for depression (Chan et al., 2007). Previous research has identified a self-positivity bias within healthy individuals; whereby, individuals are more likely to overestimate their own success compared to the success of others. Depressed individuals however, have been shown to lack such positivity bias (Alloy and Ahrens, 1987). Self-concept and self-referent bias may be of particular significance within adolescent populations. Adolescence is a sensitive period were self-concept is developing (Marcia, 1980), biases towards negative self-descriptors in this age group may impair the development of adaptive self-concepts.

Regression models predicting each outcome variable included different predictors, signaling salience of distinct cognitive bias in depression, anxiety and wellbeing. Specifically, dysfunctional attitudes emerged as a significant predictor of depression and wellbeing. Robinson and Alloy (2003) found that dysfunctional attitudes and components of rumination interacted to prospectively predict onset, number and duration of depressive episodes, in an undergraduate sample. In the present study, dysfunctional attitudes were found to be more strongly related to depression and wellbeing than anxiety. This is expected as the dysfunctional attitudes scale was developed to capture thinking styles associated with depression (Power et al., 1994). Rumination is a significant research focus and has been frequently related to emotional disorders (Young and Dietrich, 2015). Consistent with this, our findings demonstrate that rumination predicted depressive and anxiety symptoms but was less strongly associated with wellbeing.

No significant relationships between autobiographical memory recall and depression, anxiety or wellbeing were demonstrated. Previous findings have been inconsistent. Chan et al., (2007) demonstrated no significant differences in this task between individuals at high vs. low risk for depression by virtue of neuroticism. Further, Swales et al. (2010) found group differences in specificity of autobiographical memory when comparing clinical groups to controls, but that this was due to individuals within clinical groups recalling the same suicide-related memories in response to multiple cue words. However, autobiographical memory impairment in depressed groups is a robust finding with large effect sizes within adult samples (Williams et al., 1997). The above mixed findings may indicate that differences in autobiographical memory are related to symptom severity; such bias may be a scar effect rather than an antecedent risk; or , in contrast, to adult research, overgeneral memory is not a reliable cognitive marker of adolescent depression.

This study demonstrated that accuracy in identification of angry faces predicted wellbeing. This indicates that individuals with lower wellbeing were better able to recognise anger, suggesting higher sensitivity towards negative facial expressions. This is consistent with

previous research, that found that recognition accuracy of negative emotions to be associated with depressive relapse (Bouhuys et al., 1999). This finding supports the protective value of wellbeing in that, the association with a positivity bias in facial emotion identification may foster positive social interactions. However, the effect size of our finding was small and no other significant relationships between psychological outcomes and facial expressions were demonstrated. Previous research demonstrated inconsistencies that may indicate that facial emotion recognition bias is not a robust marker of mood disorders. Alternatively, there may be a task failure to detect subtle differences in interpretation. Picci and Scherf (2016) showed that adolescents were significantly better at identifying faces of their own age and hypothesised a 'dip' in facial recognition whereby there is a recalibration of the face-processing system away from caregivers towards peers. It is possible that the use of adult faces has interfered with an effect of emotion. Review of adult studies of facial emotion interpretation indicates evidence supporting an increased tendency to interpret facial expressions more negatively (Bourke, Douglas and Porter, 2010). However, as in the adolescent samples, there is discrepancy with some evidence indicating a global deficit of facial emotional processing, while some studies support a mood-congruent bias towards negative emotions in depressed groups.

A number (18%) of participants reached scores associated with clinical depression on the MFQ (Daviss et al., 2006), consistent with prevalence estimates within this age group (Avenevoli et al., 2015). This indicates the representative nature of the sample. A key strength of this study has been the assessment of wellbeing. The mental health spectrum ranges from highly disabling disorder to positive states of wellbeing. Components of wellbeing are recognised protective factors against disorders (NHS Scotland, 2016). Our results highlight the importance of cognitive biases for subjective wellbeing. Aiming to enhance positivity bias in order to boost wellbeing, potentially protecting against depression or anxiety in preventative or early intervention strategies, may be an avenue of future research. Future research may investigate the potential to develop interventions that address specific biases, relevant to individuals' experiences and symptoms, particularly in light of the importance of cognitive contributions (rumination, dysfunctional attitudes, self-referent processing and interpretation biases). Similarly, development of interventions addressing cognitive bias in order to enhance wellbeing, which is of importance for quality of life, has been associated with favourable life outcomes, including longevity (Sadler et al., 2011).

There are some limitations of this study; primarily, predictions are based on regression analyses of cross-sectional data, limiting the ability to conclude causality. Similarly, age has been demonstrated to exert non-linear development which has not been accounted for within

this study. Self-report questionnaires have been employed rather than clinical interviews. The latter was deemed less feasible due to issues of anonymity, confidentiality and the non-clinical nature of the sample. Further, although the impact of neuroticism has been analysed as distinct to cognitive factors, it is possible that neuroticism itself is influenced by mood state and biases, which may impact interpretations of the current results. Within this study, including neuroticism in initial blocks has allowed for the explanation of unique variance by cognitive biases. In future work a larger sample may be employed to allow for full mediation analysis to examine such effects. Additionally, results indicate that those who participated in cognitive tasks demonstrated significantly lower anxiety scores than those who did not complete the face-to-face cognitive tasks. This group is primarily composed of individuals from the school that withdrew from participation due to time constraints. This particular school was the only single-sex school that had been recruited for this research. As females typically demonstrate higher levels of anxiety, this may go some way to explaining the differences demonstrated here. If that were the case one may expect increases in scores of depression and neuroticism alongside increased anxiety, however no significant differences were demonstrated on these measures. It may be that higher levels of anxiety is a particular facet of this particular group. Considering this bias, specifically addressing anxiety may be a target of further investigation.

To examine a wide range of mood states, participants were recruited from community settings; therefore, results are less generalisable to clinical groups but are representative of typical adolescents. Recruiting adolescents is notoriously difficult. While the hierarchical regression models were sufficiently powered, initial identification of salient variables was underpowered and as such, variables with smaller effect sizes may not have been identified. Finally, our regression models explained approximately 50% of the variance of depression, anxiety and wellbeing, indicating that there are important factors that have not been accounted for here. For example, stressful life events and general cognitive performance would be valuable factors to include in future studies.

2.10 Conclusion

This chapter has reported an empirical study that assessed a relatively wide range of cognitive biases within a single sample using validated and standardised measures in combination with experimental paradigms. The key findings demonstrate that cognitive biases (which are potentially amendable) accounted for variability in depression, anxiety and wellbeing over and above that of known trait-like risk factors including neuroticism and attachment (which are less directly modifiable). Results highlight the importance of cognitive factors in symptoms of depression and anxiety as well as wellbeing. Furthermore, this study supports a substantial body of research implicating neuroticism, self-referential memory bias and interpretive bias in the aetiology of adolescent depression. Contributions of cognitive mechanisms, identified here, are a feasible target for behavioural modification and improvement of interventions, potentially targeting specific biases and to enhance wellbeing as a protective factor are worthy of further study.

Chapter Three - Investigating the role of attachment

3.1 Introduction

The focus of Chapter Two was to examine the contribution of cognitive bias to predictions of symptoms of depression, anxiety and wellbeing. In particular, to examine the magnitude of their predictive power above neuroticism, a heritable risk factor with a substantial genetic composition (see Chapter Two). While neuroticism has been identified as a risk factor for the development of psychopathology, environmental factors have also been considered to be of importance. The focus of this chapter is an examination of attachment in relation to cognitive biases and psychological health and wellbeing. Attachment has previously been considered to be established during early childhood experiences and is implicated in both patterns of cognition and internalizing disorders. The concept of attachment will be discussed within the introduction in relation to evidence linking it to psychological health and wellbeing with a particular focus on adolescents. Building upon the model developed in the previous Chapter, the focus of this chapter is to examine the contribution of attachment measures (parental and peer) in relation to neuroticism and cognitive biases in predicting depression, anxiety and wellbeing. An empirical study investigating these measures will be described and discussed.

Attachment Theory

Attachment theory, first proposed by Bowlby (1969, 1973, 1980), has become a prevailing theory when considering child development. According to Bowlby (1973), individuals have an innate desire to form relationships with others and the quality of these relationships is dependent on the quality of previous interpersonal interactions. Attachment theory comprises a psychological model in relation to the bonding between an individual and a primary caregiver (attachment figure). This relationship is considered to form the basis of self-concept and expectations in response to the caregivers' responsiveness and availability. As such, initial infant-caregiver relationships are of extreme importance for the emotional and social development of the child and may have long-term cognitive and behavioural implications. Bowlby (1973) posits that children develop 'internal working models' of expectations concerning the self, others and the self-in-relation-to-others, based on the attachment figure's emotional availability and responsiveness to the child's need (in Collins and Read, 1994). This internal working model of relationships developed in early childhood

is considered to be the foundation of individuals' expectations and responsiveness to future relationships.

Bowlby (1969,1982) proposed a model of attachment based upon his understanding of 'maternal deprivation', which he found to have deleterious consequences in relation to social and emotional development. Ainsworth developed a three-category understanding of attachment based on research utilising the Strange Situation paradigm. Ainsworth, Blehar, Waters and Wall (1978) demonstrated that when separated from one of their parents, approximately 60% of infants become upset and then seek out comfort from the parent upon their return. These children are considered to be 'securely attached'. However, approximately 20% of children became extremely distressed upon separation and were difficult to comfort following the return of the parent. This group has been considered 'insecure-resistant' or 'anxious-ambivalent' (frequently referred to as resistant). Approximately 20% of children demonstrate 'insecure-avoidant' attachment style, whereby they did not show significant distress at the separation and avoided contact with the parent upon return. It is theorised that children who are considered securely attached develop adaptive working models of themselves and others whereas avoidant individuals develop concepts of the self as unworthy due to rejection or unavailability of a primary caregiver. The final category, defined by Ainsworth (1971,1978), those who are considered ambivalent are thought to develop a negative self-image and exaggerated emotional responses. Hazan and Shaver (1987, 1990) described the manifestation of these attachment styles in adulthood:

Avoidant: I am somewhat uncomfortable being close to others; I find it difficult to trust them completely, difficult to depend on them. I am nervous when anyone gets too close, and often love partners want me to be more intimate than I feel comfortable being.

Anxious-Ambivalent: I find that others are reluctant to get as close as I would like. I often worry that my partner doesn't really love me or won't want to stay with me. I want to get very close to my partner, and this sometimes scares people away.

Secure: I find it relatively easy to get close to others and am comfortable depending on them. I don't often worry about being abandoned or about someone getting too close to me.

Alternative models have been proposed, for example Bartholomew and Horowitz (1991) extended Bowlby's theory, proposing two components of attachment, a model of the self and a model of other, both of which can be positive or negative. As such, based on early childhood experiences, individuals may consider themselves to be worthy or unworthy and others as trustworthy and available or unreliable and rejecting. From this model, four possible categories exist: 1) secure, (positive self, positive other); 2) preoccupied, (negative self, positive other); 3) fearful avoidant (negative self, negative other); and 4) dismissing avoidant, (positive self, negative other). A longitudinal study measured infant attachment security at 12 months of age found that attachment security in infancy was predictive of attachment security as measured at age 20 (78%). This provides support for theoretical position, that attachment is founded in infancy and remains fairly stable over time.

Measures of attachment have also been developed to capture the quality of attachments in a linear rather than categorical capacity. The Inventory of Parent and Peer Attachment was initially developed in 1987 (Armsden and Greenberg, 1987) in a sample of undergraduate students and has since been revised (Gullone and Robinson, 2005) for use in children and younger adolescents. This scale aims to capture the quality of attachment in order to assess the risk of any deleterious impact of negative attachment styles that had greater operational value and improved reliability compared to categorical style measures. Furthermore, the majority of measures that assess current attachment relationships are designed to examine romantic relationships; which may be less relevant to adolescents (e.g. Hazan and Shaver, 1987; The Relationships Questionnaire, Bartholemew and Horowitz, 1991; Experiences in Close Relationships, Brenan et al. 1998). Previous applications of this scale have demonstrated that attachment relationships with parents and peers provide somewhat similar functions and both were related to measures of adjustment, depression and wellbeing (Laible, Carlo and Raffaelli, 2000; Gullone and Robinson, 2005).

Attachment in Adolescence and the Formation of Secondary Attachments

Adolescence is a stage where individuals seek novel experiences away from attachment figures and seek increasing independence. This results in the development of interpersonal relationships with others who may become secondary (or tertiary) attachment figures, for example, romantic relationships and close friendships (Margolese, Markiewicz and Doyl, 2005). Close friendships and romantic relationships are not consistently considered as attachment relationships within the research literature. For example, Bowlby and Ainsworth

both considered that romantic relationships comprised attachment relationships, but predominantly focus on adult relationships. As attachment bonds are considered to serve an evolutionary function (i.e., supporting survival of offspring), the role of non-exclusive friendships as attachment relationships has been less discussed. Although such relationships are not motivated by sexual or reproductive systems they may still be characterised by proximity seeking and safe haven functions, which comprise key components of attachment relationships (Crowell and Waters, 1994). Some research indicates that healthy friendships are not characterised by separation distress, which is seen in other attachment relationships, however this point is debated within the literature (see Field, 1996). Allen (2007) argues that while adolescents seek comfort and support from friends, these relationships do not constitute attachment bonds (supported by Walters and Cummings 2000).

Biological mechanisms of attachment have also been researched. High endogenous oxytocin levels are associated with relationship formation and are demonstrated in in situations that have been related to attachment including peripartum and during breast feeding (e.g. Feldman 2017). Research has also exhibited increased oxytocin levels during positive social interactions between parents and children as well as between romantic partners. It has been hypothesised that oxytocin release is a mechanism promoting attachment bonding. Of importance to the current thesis is that oxytocin release has been identified as a facilitator of social bonding more generally (i.e. within non-kin relationships). Feldman (2017) posits that human attachment between parent-infant, pair-bonds, peers and conspecifics are underpinned by neural networks that are shared throughout the lifespan. Feldman discussed physiological evidence which supports the premise of attachment bonds within friendships including patterns of social reciprocity, coordination of heart rates during joint action, oxytocin release and brain-to-brain synchrony (see Feldman, 2017 for comprehensive review).

Research indicates that attachment may result in epigenetic changes that influence disorder onset. For example, in an adolescent sample (mean age=19.8), Jones-Mason, Allen, Bush and Hamilton (2016) demonstrated that methylation and genotype of SLC6A4 (promoter region of the serotonin transporter gene) was related to attachment. They found that across socioeconomic status' individuals that were classified as having unresolved attachment demonstrated lower levels of SLC64A methylation compared to those with organised or secure attachment classifications. This finding may indicate that environmental influences (e.g. abuse) may result in epigenetic alterations, thereby influencing disease risk. Consequently, attachment may be of importance in relation to gene expression and vulnerability.

Research often considers non-romantic friendship relationships as non-exclusive and without enduring commitment, which may have underestimated the importance of these relationships as in fact these can be enduring and committed. This is of particular relevance within adolescence where peer relationships and status amongst peers is highly salient. Some research has established that in adolescence particularly, a hierarchy of attachment relationships exist and that these are subject to change across development. For example, Rosenthal and Kobak (2010) found that younger adolescents (mean age = 15.9 years) placed mothers and fathers in primary and secondary positions with friends in tertiary and quaternary positions and that older adolescents (mean age = 19.3 years) demonstrated a marked shift in that most participants placed friends in secondary positions, thereby identifying them higher in the attachment hierarchy than biological fathers. Rosenthal and Kobak (2010) also found that placing friends in primary or secondary positions was related to an increased risk for externalising symptoms in males and females, and was related to increased risk of internalising symptoms in females. This may indicate that preference for support from friends rather than parents represents a maladaptive strategy that is related to adverse outcomes. Bowlby (1969, 1982). argued that adolescents demonstrate individual differences in attachment hierarchies in that: some separate from parents, while others remain very attached and are unwilling to direct attachments towards others (Rosenthal and Kobak (2010) concluded that disengagement from parents during early adolescence may be maladaptive resulting in 'premature autonomy' related to problem behaviour. However, it may be that in these cases individuals seek increased support from peers as attachment needs are unmet by parental attachment figures, although this cannot be inferred from the results.

While some research has related negative outcomes with strong peer attachment in adolescents, others have demonstrated that perceived social support from peers acts as a protective factor for adolescent mental health in individuals that are impacted negatively by parental behaviour such as those affected by domestic violence (Levendosky, Huth-Bocks and Semel, 2010). One implication of this finding is that it may be that positive secondary attachment relationships with peers may attenuate the effects of primary attachments. Hazan and Zeifman (1994) found that adolescents preferred their friends as sources of comfort and emotional support, but parents continued to be preferred as sources of security. Similarly, Muris et al., (2001) found that among adolescents, parents were scored more highly than peers on a measure of trust in relation to attachment figures, indicating that their parental attachment was of relatively greater importance in relation to trust. However, on the communication subscale, peers were scored more highly than parents, reflecting the shift away

from parents and increased importance of social relationships during this period (Muris et al., 2001).

Furthermore, social status, quality of friendships and quality of romantic relationships was found to protect against anxiety while social status amongst peers was protective against depression symptoms (La Greca and Harrison, 2010). Furthermore, La Greca and Harrison (2010) showed that negative quality friendships and romantic relationships predicted depression symptoms in adolescents. This highlights the importance of such relationships (which may comprise attachment bonds), at this age for the health and wellbeing of individuals and suggests that various aspects of social relationships contribute to distress.

Attachment, Cognition and Disorder

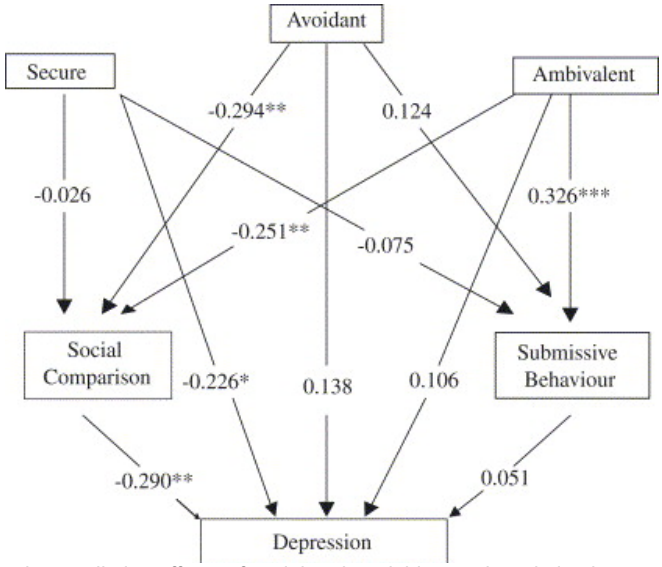
Attachment theory predicts that negative internal working models of the self and others, associated with early caregiving experiences, are related to the patterns of cognition observed in depression. Ciccetti, Cummings, Greenberg and Marvin and (1990) hypothesize that insecure attachments contribute to internalized feelings of low security. Insecurely attached individuals are therefore likely to interpret negative interpersonal events as a reflection of their unworthiness, and tend to have fewer psychological resources for coping during times of stress, which all contributes to depressed mood. This association between depressogenic cognitions and poor attachment seems to make intuitive theoretical sense, particularly when considering the prediction that insecurely attached individuals demonstrate negative interpretations and expectations of social relationships. This bares conceptual similarity to Beck's (1967) negative triad of depression which specifies individuals hold a negative internal model of the self, the world and others.

Depression, anxiety and wellbeing

Poor quality attachment is often considered to constitute a risk factor for the development of psychopathology, while, good quality or secure attachments may be considered a protective factor of psychological health. A longitudinal study with a large and representative Norwegian sample, indicated that insecure parental attachment was associated with the presence of depression between the ages of 15-20. In particular, maternal attachment was most strongly associated with the group that 'remained depressed' across the follow-up period

(Agerup, Lydersen, Wallander, Sund; 2015). However, peer attachment was not significantly associated with depression variables.

Irons and Gilbert (2005, see Figure 3.1) found higher scores of secure attachment to be predictive with anxiety and depression in adolescents (mean age=14.63), lower attachment security predicted depression. Raja, McGee and Stanton (1992) proposed parental attachment as a critical factor for psychological health in adolescence supported by data from a cohort sample (N=935, data collected at age 15). This study demonstrated that high levels of wellbeing were most strongly associated with parental rather than peer attachment. Post-hoc testing indicated that the highest depression scores were most strongly related to low-quality parent and high-quality peer attachments (Raja et al, 1992). This supports the assertion that good quality peer attachments did not compensate for low quality parental attachments.



Mediation analysis showing mediation effects of social rank variables on the relation between attachment style and depression. *p<.05, **p<.01 and ***p<.001.

Figure 3.1: Irons and Gilbert (2005) mediation analysis depicting the relationship between attachment style and depression

As discussed in previous chapters, neuroticism is a key heritable risk factor for depression and anxiety. Debate exists surrounding the nature of the relationship between attachment and neuroticism, which has been implicated as a risk for psychological disorders (see Chapters One and Two). A review of the relationship between attachment and neuroticism found that secure attachment was negatively related neuroticism, anxious attachment was positively related to neuroticism (r=.42; p<0.001); and avoidant attachment demonstrated a positive correlation with neuroticism (r=.14; <0.001). There was however, less consistency between the reviewed studies when looking at the relationship between avoidant attachment and neuroticism (Noftle and Shaver, 2006).

Although relationships with neuroticism have been demonstrated, some propose that the high correlations between attachment style and personality traits indicate poor discriminant validity of the construct of attachment. Crawford, Shaver and Goldsmith (2007) discussed this issue in detail and identified that the relationship between neuroticism and attachment style is complex and is influenced by multiple factors. In a twin study, Crawford, Livesley, Jang, Shaver, Cohen and Ganiban (2007) identified shared genetic effects between pathological personality dimensions and attachment styles. Although this study did not directly measure neuroticism, implications for this area remain. Authors posit that neuroticism may provide an underlying genetic basis for anxious attachment as well as emotional dysregulation, thereby predicting the overlap (Crawford et al., 2007). It may be that individuals are impacted by the availability of attachment figures to varying degrees depending upon their predisposition to experience anxious attachment (Crawford et al., 2007). Neuroticism is considered to increase the susceptibility to disorders and similarly, may lead to greater activation of the attachment system. The implication of this is that individuals with high neuroticism are more likely to seek greater comfort from attachment figures and be more concerned about the availability of attachment figures. As such, attachment and neuroticism have been considered to be related yet distinct concepts.

Cognition

The proposed role of cognitive biases in the aetiology of disorders has been discussed in previous chapters (see Chapter One and Two). The relationship between attachment and cognitive bias has rarely been addressed by previous research. Attachment style has been implicated in the relationship between cognitive bias and emotional disorder. Cognitive styles, such as dysfunctional attitudes and rumination, have been implicated in the relationship between attachment and depressive disorder. For example, Reinecke and Rodgers (2001) demonstrated that dysfunctional attitudes partially mediated the relationship between attachment style and depression in a sample of clinically depressed adults. Similarly, rumination has also been proposed to serve a mediating function between quality of attachment (assessed using the Inventory of Parent and Peer Attachment) and depression in a non-clinical sample of adolescents (mean age 14.3 years), thereby demonstrating the complex relationship between cognition, attachment and health and wellbeing.

Both attachment and cognitive biases are frequently associated with emotional disorders yet the two are infrequently examined in relation to each other. The importance of cognitive biases

in relation to psychological symptoms has been supported by results from Chapter Two. Barrett and Holmes (2001) demonstrated significant relationships between attachment styles and responding to an ambiguous scenarios task in older adolescents (mean age=19.05). Insecure attachment with parents and peers was predictive of negative interpretation bias (explaining approximately 55% of the variance of interpretation bias). Furthermore, parental attachment accounted for a large portion (40%) of this variation (Barrett and Holmes, 2001). If emotional processing biases constitute vulnerability to depression, this is of particular importance in an adolescent context considering the prevalence of psychological illness and the changing nature of social and attachment relationships.

Attachment theory makes predictions about interpersonal relationships and internal cognition, which may increase risk for disorder. As attachment is formed during early life experiences and as such may be less responsive to subsequent modification, making it a complex target for intervention. The cognitive style associated with low quality attachments and depression may however, predict other negative biases of cognition which may in turn precipitate depression. These cognitive mechanisms may be more responsive to treatment which may indirectly modify the impact of attachment style developed early in life. Furthermore, intervention during adolescence may positively impact internal working models surrounding expectations and attachments related to current and future relationships. As such good quality attachments potentially promote positive interpersonal functioning, with implications for the maintenance of healthy social relationships and wellbeing.

3.2 Rationale

Considering that attachment security is considered to be related to psychological problems and adolescence is a time where attachment relationships may undergo modification as well as being a high-risk period for the onset of psychiatric disorders, the relationship between these variables is worthy of study. However, relatively few studies address current attachments (e.g. peer) in relation to adolescent mental health, which is considered a particularly crucial stage in the onset of psychological disorders and a critical developmental period.

3.3 Study Aims and Hypotheses

The aims of this study are therefore to build upon the model established in Chapter Two, to consider the additional contribution of parental and peer attachment. It was demonstrated that cognitive biases (self-referential, and interpretation bias) and cognitive styles (rumination and dysfunctional attitudes) accounted for variance in measures of depression, anxiety and wellbeing independently of neuroticism. This chapter aims to include measures of parental and peer attachment to evaluate the contribution of attachment in these relationships and to consider the impact of attachment on cognitive biases and symptoms of depression, anxiety and wellbeing.

Based on previous research, it was predicted that parental and peer attachment would be significant predictors of depression, anxiety and wellbeing. Considering that parent and peer attachment may serve similar functions, and the theoretical prediction that parental attachment establishes attachment in future relationships, it may be that parental attachment demonstrates greater salience than peer attachment in relation to both symptoms of outcome measures and cognitive biases. It is expected that attachment quality measures will increase the amount of variance that is explained, and that this increase will be unique from measures of neuroticism and cognitive bias.

Based on findings from Chapter Two, it was hypothesised that cognitive biases of rumination, dysfunctional attitudes, ambiguous scenarios and self-referential memory will significantly predict symptoms and that autobiographical memory and facial emotional expression recognition will not demonstrate predictive significance. It was also hypothesised that interpretation and self-referential recall bias will demonstrate strong relationships with attachment.

3.4 Methodology

Participants

This study is based on the same sample as within the previous chapter (see Chapter Two for full details). Adolescents aged 12-18 years (mean=14.9, S.D. = 1.52, N=99) from Scottish secondary schools (58.6% female and 70.3% White British), were recruited. A power calculation determined that the current sample size allowed for up to six predictors in a regression model with 80% power to detect a medium effect size.

Measures and Procedure

Data for this study were collected alongside that of the previous chapter (Chapter 2), as such full details of methodology and procedure will not be repeated here. In addition to measures described in the previous chapter, The Inventory of Parent and Peer Attachment (IPPA; Gullone and Robinson, 2005; see below) was employed to capture the quality of attachment in relation to primary caregivers and peers.

The Inventory of Parent and Peer Attachment – Revised, for children (IPPA; Gullone and Robinson, 2005)

Gullone and Robinson's (2005) revision of the IPPA extends its use to younger adolescents (9 – 11) while remaining applicable to older adolescents (14+). This instrument assesses attachment in relation to parents and peers. Both subscales consist of 28 items and assess elements of trust, communication and alienation. Internal consistency in this sample was high for both parent (Chronbach's α=.94), and peer (Chronbach's α=.90) subscales. In keeping with other measures and the parametric nature of statistical analysis employed within this study, total scores were intended to be employed. However, due to missing data mean scores were used (see section 3.7 below for more detail).

As full details of other measures and protocol are described in Chapter Two they will only be briefly described here. The IPPA was included within online collection of data alongside other psychological variables: the Mood and Feelings Questionnaire (MFQ; Angold and Costello, 1987); Spence Children's Anxiety Scale (SCAS; Spence, 1988); BBC Well-being Scale (BBC; Kinderman, Schwannauer, Pontin and Tai; 2011); Eysenck Personality Questionnaire-Neuroticism subscale (EPQ-N; Eysenck et al., 1985); The Ruminative Response Styles scale (RRS; Treynor et al., 2003); Dysfunctional Attitudes Scale: 24 Item Version (DAS-24; Power et al., 1994). See Chapter Two for full descriptions.

Measures of cognitive bias

Self-Reference Recall Task, (Kelvin et al., 1999); Autobiographical Memory Test (AMT; Williams and Broadbent, 1986); Ambiguous Scenarios Task for Depression in Adolescents (AST; Orchard Pass and Reynolds, 2016) and a Facial Expression Recognition Task (Chan et al., personal communication), were conducted face-to-face.

3.5 Statistical Analysis

Due to a procedural error, item 13 from the BBC Well-being Scale was missing at random for the majority of participants. Additionally, one item from the parent subscale of the IPPA was also missing at random. Methods of imputing data (imputing the mean and multiple imputation) were investigated and considered. An attempt to contact the authors was made, with no response, in relation to this issue. However, upon advice from statisticians, this item was excluded from calculations and the mean rather than total score was employed for further analysis. The reliability of this scale remained high (α=.95).

Tests of normality were conducted to verify the appropriateness of the chosen analysis which indicated assumptions of normality were met. Despite variables being highly correlated (see results), collinearity and tolerance statistics were within accepted limits (maximum VIF=1.50 and lowest tolerance =0.61; Field, 2009, p 297).

False discovery rate (FDR) correction for multiple comparisons, calculated using R version 3.2.5, was applied to significance values of Pearson's correlations examining relationships between variables.

Replicating the methods employed in Chapter Two to identify salient variables for predictions, backwards elimination regression was conducted for each of MFQ, SCAS and BBC. All variables (including age and gender) were entered (see Table 3.2 for a list of variables included) in a backwards elimination regression with the removal criteria of F>=0.10. Table 2 depicts the resulting final model for each outcome variable.

To assess the unique contribution of each variable, hierarchical regression was employed in order to assess the increase of r^2 at each step. Variables were entered based on their identification from the previous regression models. In each model, age and gender were included as initial block. As neuroticism is considered to be a partly heritable risk factor, and was highlighted as salient for each outcome variable, it was included as the second step variable in each regression. Regression coefficients and standardized beta-values were examined for each regression model.

As this chapter's focus is attachment, and its relative contribution to depression, anxiety and wellbeing in relation to both neuroticism and cognitive bias, the attachment variables

identified by the removal regression were entered in the third step. The fourth step included the addition of selected cognitive bias variables.

3.6 Results

Descriptive statistics are presented in Table 3.1 and Pearson's correlation analysis is presented in Figure 3.2 and Table 3.2. The strength of associations has also been identified by colour based on the key below the image.

Table 3.1: Descriptive Statistics

Measure	Mean	S.D	Minimum	Maximum
Age	14.94	1.52	13	18
MFQ (Depression)	15.26	12.88	0	57
SCAS (Anxiety)	27.59	17.09	2	95
BBC (Wellbeing)	68.56	12.87	35	92
Neuroticism	6.24	3.41	0	12
IPPA Parent	42.95	10.51	27	73
IPPA Peer	39.66	8.11	25	64
RRS (Rumination)	21.11	6.41	10	40
DAS (Dysfunctional Attitudes Scale)	91.55	17.45	51	168
AMT: Autobiographical Memory Task (Specific)	7.36	1.95	2	10
AMT: Autobiographical Memory Task (Overgeneral)	2.04	1.66	0	7
AST: Ambiguous Scenarios Task Bias	1.33	5.76	-15	15
SRR: Self-Referential Recall 'Me'	0.33	0.29	-0.50	1
SRR: Self-Referential Recall 'Not Me'	-0.23	0.19	-0.56	0.50
Facial Expression Recognition: Anger	0.57	0.15	0.23	0.88
Facial Expression Recognition: Disgust	0.43	0.15	0.05	0.73
Facial Expression Recognition: Fear	0.60	0.12	0.25	0.80
Facial Expression Recognition: Happy	0.76	0.08	0.48	0.93
Facial Expression Recognition: Sad	0.67	0.20	0.15	0.95

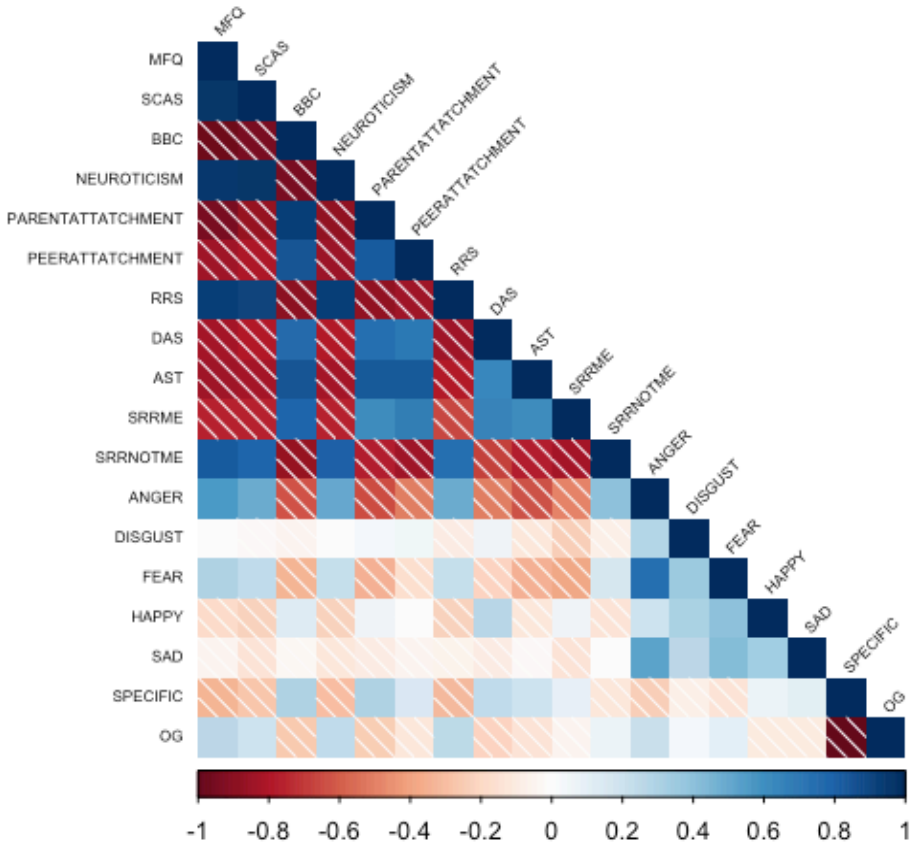


Figure 3.2:Heat Map of Strength of Association based on Pearson's Correlations

*Table 3.2: Pearson's Correlation (Age and Gender Controlled, FDR p-value correction applied) **p<0.001; *p<0.05, n=99*

	MFQ	SCAS	BBC	NEUROTICISM	IPPA PARENT	IPPA PEER	RRS	DAS	AST BIAS	SRR (' ME') BIAS	SRR (' NOT ME') BIAS	ANGER	DISGUST	FEAR	HAPPY	SAD	SPECIFIC MEMORIES	OVERGENERAL MEMORIES
MFQ	-																	
SCAS	0.71**	-																
BBC	-0.73**	-0.55**	-															
NEUROTICISM	0.63**	0.71**	-0.57**	-														
IPPA PARENT	0.65**	0.46**	-0.65**	0.45**	-													
IPPA PEER	0.35**	0.22	-0.48**	0.38**	0.38**	-												
RRS	0.58**	0.57**	-0.46**	0.56**	0.43**	0.34**	-											
DAS	-0.51**	-0.48**	0.50**	-0.54**	-0.42**	-0.34**	-0.34**	-										
AST	-0.41**	-0.41**	0.48**	-0.39**	-0.42**	-0.43**	-0.3	0.21	-									
SRR ('ME')	-0.35**	-0.32*	0.44**	-0.31*	-0.12	-0.25	-0.18	0.31*	0.15	-								
SRR ('NOT ME')	0.42**	0.34**	-0.48**	0.36**	0.31	0.42**	0.31*	-0.26	-0.35	-0.47	-							
ANGER	0.21	0.17	-0.27	0.15	0.27	0.17	0.15	-0.17	-0.33**	-0.11	0.05	-						
DISGUST	0.01	-0.08	0.07	-0.04	-0.14	-0.13	-0.07	0	-0.05	-0.14	-0.12	0.19	-					
FEAR	0.14	0.08	-0.02	0.05	0.06	-0.11	0.13	0.01	-0.08	-0.12	0.03	0.46**	0.22	-				
HAPPY	-0.10	-0.15	0.15	-0.20	-0.02	0.05	-0.10	0.12	-0.14	0.03	-0.05	0.15	0.12	0.31*	-			
SAD	-0.07	-0.15	0.04	-0.13	0.08	0.03	-0.05	-0.03	0.06	-0.06	0.08	0.35**	0.19	0.22	0.22	-		
SPECIFIC	-0.06	0	0.04	-0.01	0.01	0.05	0.04	0.04	0.03	-0.04	0.11	0.01	-0.12	0	0.04	0.12	-	
OG	0.01	-0.07	-0.03	-0.01	-0.01	-0.03	-0.09	-0.08	0.01	0.08	-0.10	-0.03	0.06	-0.02	-0.08	-0.10	-0.90**	-

Backwards Elimination Regression

The variables identified as salient in predicting MFQ were neuroticism, parental attachment (IPPA), rumination (RRS), and self-referential recall 'me' bias; in predicting SCAS were neuroticism, peer attachment (IPPA), rumination (RRS) and ambiguous scenarios task (AST) bias; and finally, in predicting BBC were neuroticism, parental attachment (IPPA), ambiguous scenarios task (AST) bias, and self-referential recall 'me' bias. See Table 3.3 for full results. Figure 3.1 depicts correlation analysis to demonstrate the strength of association between variables.

Table 3.3: Backwards Elimination Regression Results

Backwards Removal Regression Final Model (Included predictors)	**r**	**r^2**	**F**	**Sig (p)**
MFQ (Neuroticism, Parental Attachment, Rumination, SRR 'Me')	0.79	0.63	38.54 (4,91)	<0.001
SCAS (Neuroticism, peer attachment, rumination, Ambiguous Scenarios Task, SRR 'Me')	0.79	0.62	29.93 (5,90)	<0.001
BBC (Neuroticism, Parental attachment, Ambiguous Scenarios Task, SRR 'Me')	0.79	0.63	38.34 (4,91)	<0.001

Hierarchical Regressions

In predictions of MFQ, measuring depression symptoms, steps 2, 3 and 4 demonstrated a significant ($p<0.01$) increase in predicted r^2. This indicates that the inclusion of neuroticism, attachment and cognitive bias variables each increased the explained variance. The final step (including: age, gender, neuroticism, parental attachment, rumination and self-referential 'me' bias variables) demonstrated $r^2=0.64$; $F(6,92)=26.65$, $p<0.001$. Neuroticism ($\beta=0.28$), parental attachment ($\beta=-0.43$), rumination ($\beta=0.20$) and self-referential 'me' ($\beta=-0.17$) variables each demonstrated significant beta values ($p<0.05$). See Table 3.4 for full results.

Table 3.4: Hierarchical Regression Predicting Mood and Feelings Questionnaire (MFQ), n=99

		r	r^2	Adj r^2	F-value (df)	Unstandardized Coefficients B	SEB	β	t-value	Δr^2	ΔF (df)
MFQ ***Step 1***		0.19	0.04	0.02	1.77 (2,96)					0.04	1.77 (2,96)
	Constant					-4.41	12.64		-0.35		
	Gender					1.19	0.83	0.14	1.43		
	Age					3.31	2.61	0.13	1.27		
Step 2		0.64	0.41	0.39	22.22** (3,95)					0.38	60.90 ** (1,95)
	Constant					-8.74	9.93		-0.88		
	Age					0.68	0.66	0.08	1.03		
	Gender					-2.74	2.19	-0.11	-1.25		
	Neuroticism					2.49	0.32	0.66	7.80 **		
Step 3		0.76	0.58	0.56	32.95* (4,94)					0.17	38.70 ** (1,94)
	Constant					33.95	10.85		3.13*		
	Age					0.69	0.56	0.08	1.24		
	Gender					-1.26	1.87	-0.05	-0.68		
	Neuroticism					1.64	0.30	0.44	5.45**		
	IPPA Parent					-15.75	2.53	-0.46	-6.22**		
Step 4		0.79	0.64	0.61	26.65* (6,92)					0.05	6.43 * (2,92)
	Constant					28.37	11.17		2.54*		
	Age					0.69	0.53	0.08	1.31		
	Gender					-0.85	1.77	-0.03	-0.48		
	Neuroticism					1.05	0.33	0.28	3.18*		
	IPPA Parent					-14.53	2.46	-0.43	-5.90**		
	RRS					0.40	0.16	0.20	2.56*		
	SRR 'Me'					-7.45	2.98	-0.17	-2.50*		

In predictions of SCAS, measuring anxiety symptoms, steps 2 and 4 demonstrated a significant ($p<0.01$) increase in predicted r^2. This indicates that the inclusion of neuroticism and cognitive bias variables each increased the explained variance, but inclusion of peer attachment did not. The final step (including: age, gender, neuroticism, peer attachment, rumination, self-referential 'me' bias and ambiguous scenarios task bias variables) demonstrated $r^2=0.62$; $F(7,91)=20.99$, $p<0.001$. Neuroticism ($\beta=0.54$), peer attachment ($\beta=0.18$), rumination ($\beta=0.23$) and ambiguous scenarios task bias ($\beta=-0.19$) variables each demonstrated significant beta values ($p<0.05$). Beta values of self-referential 'me' ($\beta=-0.12$) did not demonstrate significance. See Table 3.5 for full results.

Table 3.5: Hierarchical Regression Predicting Spence Children's Anxiety Scale (SCAS), n=99

		r	r^2	Adj r^2	F-value (df)	Unstandardized Coefficients B	SEB	β	t-value	Δr^2	ΔF (df)
SCAS		0.29	0.08	0.06	4.34* (2,96)					0.08	4.34* (2,96)
Step 1	Constant					8.45	16.36		0.52		
	Age					0.91	1.08	0.08	0.83		
	Gender					9.62	3.38	0.28	2.85*		
Step 2		0.73	0.54	0.52	36.93 (3,95) **					0.46	93.71 (1,95)**
	Constant					2.14	11.69		0.18		
	Age					0.16	0.77	0.01	0.20		
	Gender					0.80	2.58	0.02	0.31		
	Neuroticism					3.63	0.38	0.72	9.68**		
Step 3		0.74	0.54	0.52	27.74(4,94)**					0.00	0.43 (1,94)
	Constant					-5.04	14.84		-0.34		
	Age					0.07	0.78	0.01	0.09		
	Gender					0.56	2.60	0.02	0.21		
	Neuroticism					3.76	0.41	0.75	9.18**		
	IPPA Peer					3.20	4.06	0.06	0.79		
Step 4		0.79	0.62	0.59	20.99(7,91)**					0.08	6.04 * (3,91)
	Constant					-25.46	14.89		-1.711		
	Age					0.18	0.73	0.02	0.246		
	Gender					1.55	2.43	0.05	0.641		
	Neuroticism					2.73	0.45	0.54	6.00**		
	IPPA Peer					9.29	4.07	0.18	2.28*		
	RRS					0.61	0.21	0.23	2.89*		
	SRR 'Me'					-7.14	4.12	-0.12	-1.73		
	AST					-0.56	0.22	-0.19	-2.49*		

In predictions of BBC, measuring wellbeing, steps 2, 3 and 4 demonstrated a significant ($p<0.001$) increase in predicted r^2. This indicates that the inclusion of neuroticism and cognitive bias variables each increased the explained variance, but inclusion of peer attachment did not. The final step (including: age, gender, neuroticism, parental attachment, self-referential 'me' bias and ambiguous scenarios task bias variables) demonstrated $r^2=0.63$; $F(76,92)=26.29$, $p<0.001$. Neuroticism ($\beta=-0.21$), parental attachment ($\beta=0.45$), self-referential 'me' bias ($\beta=0.29$) and ambiguous scenarios task bias ($\beta=0.15$) each demonstrated significant beta values ($p<0.05$). See Table 3.6 for full results.

Table 3.6: Hierarchical Regression Results Predicting BBC Subjective Wellbeing Scale (BBC), n=99

		r	r^2	Adj r^2	F-value (df)	Unstandardized Coefficients B	SEB	β	t-value	Δr^2	ΔF (df)
BBC		0.24	0.06	0.04	2.80(2,96)					0.06	2.80 (2,96)
Step 1	Constant					89.43	12.50		7.15**		
	Age					-1.21	0.82	-0.15	-1.46		
	Gender					-4.91	2.58	-0.19	-1.90*		
Step 2		0.59	0.35	0.33	17.01** (3,95)						
	Constant					93.25	10.45		8.93**	0.29	42.99 ** (1,95)
	Age					-0.75	0.69	-0.09	-1.09		
	Gender					0.43	2.30	0.02	0.19		
	Neuroticism					-2.20	0.34	-0.58	-6.56**		
Step 3		0.73	0.54	0.52	27.18** (4,94)					0.19	37.87 ** (1,94)
	Constant					48.72	11.45		4.26**		
	Age					-0.77	0.59	-0.09	-1.31		
	Gender					-1.12	1.97	-0.04	-0.56		
	Neuroticism					-1.32	0.32	-0.35	-4.14**		
	IPPA Parent					16.43	2.67	0.49	6.15**		
Step 4		0.80	0.63	0.61	26.29** (6,92)					0.10	11.90** (2,92)
	Constant					45.65	10.77		4.24**		
	Age					-0.88	0.53	-0.11	-1.64		
	Gender					-1.52	1.78	-0.06	-0.85		
	Neuroticism					-0.78	0.31	-0.21	-2.54*		
	IPPA Parent					15.15	2.53	0.45	5.99**		
	SRR 'Me'					12.96	3.00	0.29	4.33**		
	AST					0.33	0.16	0.15	2.05*		

3.7 Discussion

Results demonstrate that an increased proportion of symptoms of depression and wellbeing were predicted by parental attachment and cognitive bias variables. In models of depression and wellbeing, parental attachment was a significant predictor and increased the explained variance by 17-19%. In the regression model predicting anxiety, peer attachment rather than parental attachment was included based on the initial elimination regression. The final step of this hierarchical regression indicated that peer attachment significantly impacted the regression slope (β=0.18). However, its inclusion at step three (adding in peer attachment) failed to demonstrate a significant increase of explained variance. This suggests that overall parental attachment plays a stronger role than peer attachment in adolescent mental health and wellbeing.

The above results support the hypothesis that parental attachment would be a stronger predictor than peer attachment. However, there was a distinction between the salience of salience parental and peer attachment in relation to symptoms. Parental attachment was identified as the most salient variable in relation to depression and wellbeing. Whereas in relation to anxiety peer attachment was identified to be more relevant, although it did not significantly improve predictions. It may be that peer attachment overlapped with another variable (potentially neuroticism) and thus failed to contribute unique variance to the model. This is important as it may indicate that attachment relationships are more important to the development of depression and protection of wellbeing compared to the development of anxiety. This would be in line with previous research that identifies close relationships and/or overlaps of attachment and neuroticism (e.g. Crawford et al., 2007). This is similar to the findings of Crawford et al., (2007), who report correlations between attachment and neuroticism to be approximately r^2=0.16-0.25. Furthermore, Crawford et al., (2007) demonstrated that the relationship between attachment and neuroticism was not linear. One measure of attachment (avoidant type) was found to moderate the relationship between a second measure of attachment (anxious type) and neuroticism (Crawford et al., 2007). However, like the present study, the study by Crawford et al., (2007) was cross-sectional and therefore, directionality cannot be inferred.

Somewhat unexpectedly, parental attachment variables had a greater impact on models of depression and wellbeing than neuroticism. Although both were significant, in final models of MFQ and BBC, beta-values of parental attachment were larger than those of neuroticism.

Negative correlations between attachment variables and neuroticism were r^2= 0.18. This indicates that lower quality of parental attachment was predictive of higher depression and lower wellbeing and that parental attachment was superior in predicting this compared to neuroticism. However, it is important to consider this finding with caution due to the close relationship between neuroticism and attachment.

In final models, each regression predicted a similar amount of the variance of each outcome variable, 62-64%. This prediction explains a greater amount of variance than the previous chapter which did not include attachment variables; final models in Chapter Two accounted for 52-59% of variance of depression, anxiety and wellbeing variables. This is of importance as a high proportion of symptoms has been explained by these models, particularly considering that limited research has addressed these variables within single studies in adolescence. This is of importance as a high proportion of symptoms has been explained by these models, particularly considering that limited research has addressed these variables within single studies in adolescence.

Supporting hypotheses, predictions of MFQ and BBC demonstrated a significant increase of explained variance when adding parental attachment, as well as, a further significant increase in explained variance when including cognitive bias variables (5% and 10% respectively). Although attachment variables did not increase the explained variance of anxiety symptoms, cognitive biases contributed unique variance, accounting for an 8% increase of the explained variance. This indicates that while an increase of variance was accounted for by attachment variables, cognitive bias variables continued to contribute unique variance. Chapter Two identified that cognitive bias variables contributed to the variance of depression (12%), anxiety (5%) and wellbeing (19%). Comparing the difference in these estimates between chapters indicates that there may be some overlap between the effect of attachment and cognitive bias. As such, this Chapter has identified that some of the variability accounted for by cognitive bias in the previous chapter may be attributable to attachment variables. However, there remains a significant impact of cognitive biases on the symptoms of depression anxiety and wellbeing, supporting the role of cognitive biases in disorder aetiology.

In relation to cognitive bias, parental and peer attachment security demonstrated significant correlations with measures of rumination, dysfunctional attitudes, and interpretation biases of ambiguous scenarios. Supporting hypotheses and previous findings such as those of relationships between attachment and interpretation bias (Crawford et al., 2007). Associations

were in the expected direction, in that greater security of attachments was associated with reduced rumination, fewer dysfunctional attitudes and reduced positivity bias in the interpretation of ambiguous scenarios. In relation to self-referential recall memory bias, peer attachment was significantly related to both endorsed and non-endorsed recall bias; whereas, parental attachment was only significantly correlated to non-endorsed recall bias. Furthermore, the directionality of association was as expected and mirrored that of the relationships with depression, anxiety and wellbeing variables. This means that lower quality attachment was related to more negative biases in recall of words endorsed as self-referential and that lower quality attachment was related to greater positivity of bias in relation to words not endorsed as self-referential. This again highlights the importance of self-concept in relation to health and wellbeing. As discussed in previous chapters, results suggest that this negative bias is specific to concepts related to the self rather than a globally negative bias (see Chapter Two). The formation of attachment relationships may be involved in this process. Attachment theory identifies that individuals' understanding of their worth is developed in relation to the availability responsiveness of attachment figures (Bowlby, 1973). This may impact potential biases held by individuals in relation to development of their self-concepts and therefore facilitating positive or negative biases.

Similarly, attachment may elicit effects in relation to interpretation bias. Early attachment relationships are considered to develop understanding of one's relative worth in relation to others and this is considered to form a basis for future relationships (Bowlby, 1973). Consequently, individuals may employ attachment models during evaluation of everyday scenarios, thereby predisposing one to positive or negative biases within the realm of interpretation biases. It may be possible to identify individuals who may be of increased risk due to lower quality attachments and intervene with early interventions to modify cognitive biases in hopes to protect against disorder onset.

It was expected (based on findings of Chapter Two), that correlations between attachment and autobiographical memory task bias would be low and likely non-significant. Similarly, that biases of recognition of facial emotions would be non-significant, other than in relation to angry expressions. As predicted both parental attachment and peer attachment failed to demonstrate significant correlations with specific or overgeneral memory bias (and these variables were excluded in elimination regressions). Recognition bias of anger demonstrated significant correlations with parental but not peer attachment. Attachment relationships may salient in relation to the processing of interpersonal stimuli, such as faces. While attachment

has not been frequently studied in relation to information processing, Niedenthal, Brauer, Robin and Innes-Ker (2002) demonstrated a distinction in the perception of facial emotional expressions between groups of securely and insecurely attached adults. Under conditions of distress, securely attached adults were quicker to identify the offset of morphing negative to neutral facial expressions, while insecurely attached adults identified negative facial expressions as occurring for longer (Niedenthal et al., 2002). Faces convey social information relevant to the maintenance of social relationships (Marsh, Ambady and Kleck, 2005). It may be posited that attachment styles may impact individuals' sensitivity to interpersonal cues of the emotional availability of others, thereby demonstrating differences in the processing of facial expressions. In this study, the correlation coefficient of the relationship between angry facial expressions and attachment was low, predicting only 7% of scores. As discussed in Chapter Two, previous examination of facial processing biases have been inconsistent in relation to adolescent populations. There may be a failure of this specific task to identify subtle effects of facial processing biases. As discussed previously, there is also a potential issue in employing adult faces as stimuli within adolescent samples due to a hypothesised dip in facial recognition during the adolescent period (Picci and Scherf, 2016; see Chapter Two).

In models of SCAS, peer attachment demonstrated a significant impact on the regression slope with (β=0.18). It should be noted that the relationship here is positive; increased SCAS score was unexpectedly predicted by an increase in quality of peer attachment (as measured by the IPPA-R peer subscale). However, correlation analysis indicates a negative relationship between the two variables (higher anxiety with lower quality peer attachment). This may indicate that the negative relationship demonstrated in correlational analysis was influenced by a third variable. That the effect changes within regression models suggests that the influencing variable has been included within the model, resulting in peer attachment no longer accounting for unique variance. One possibility is that neuroticism exerts this effect on the relationship between peer attachment and anxiety. As the regression model included neuroticism prior to the inclusion of peer attachment, it may be that neuroticism mediates the impact of peer attachment on anxiety (or vice versa). Previous work as well as findings from Chapter Two and the current chapter identify neuroticism as exerting strong effects on anxious symptomology (β=0.75 in the current results). If this is the case, personality traits may influence the development of attachment styles, particularly in relation to peer relationships; or, mediate the impact of attachment styles on anxiety.

Limitations

Alongside limitations discussed in Chapter Two in relation to cross-sectional analysis, non-linear effects, self-report data and the potential interdependent relationship between neuroticism and depression, anxiety and wellbeing, there are some further limitations associated with this study. Issues identifying causality are of particular importance here in terms of the relationship between attachment and neuroticism. Conducting full moderation analysis may increase understanding of the relationship between variables, particularly between that of attachment and neuroticism. Longitudinal analysis may also be employed to assess causality. Similarly employing measures that capture attachment style based on retrospective assessments of relationships with parents, unlike the measure employed here which measured quality of current attachments. As discussed previously, there are also limitations due to the nature of the sample. While recruitment from community settings has been beneficial in terms of increasing understanding of the spectrum of depression, anxiety and wellbeing, it may be a limitation in terms of increasing understanding of clinical disorders. Finally, the issue of power remains, final hierarchical models were sufficiently powered; however, initial elimination were underpowered and as such, variables exerting small effects may not have been identified.

3.8 Conclusion

This chapter has strongly implicated the role of parental attachment in symptoms of depression and wellbeing. Parental attachment was identified as being of greater salience to depression and wellbeing, and contributed significantly to predictions. Peer attachment was initially identified as of more relevance to anxious symptomology, inclusion of peer attachment however, did not significantly increase predictions of anxiety. Some overlap between neuroticism and attachment was demonstrated. Despite the large roles of both neuroticism and attachment, this study indicated that cognitive biases continued to contribute unique variance in explaining depression, anxiety and wellbeing. In conclusion, attachment is identified as a key mechanism and early intervention work may attempt to improve attachment security in order to minimise depression. Additionally, cognitive mechanisms warrant further study particularly in relation to potential modification to improve wellbeing and in preventative intervention to protect against depression and anxiety.

Chapter Four - The Stress-Response System: A Potential Biomarker?

4.1 Introduction

The previous two chapters focused on cognitive contributions to psychological health and wellbeing. As part of a holistic approach to address the impacts of emotion and emotional disorders on individuals, this chapter aims to examine physiological mechanisms and their relationship to psychological constructs. Associations between physical and psychological health have been observed and there appears to be a disproportionate impact of physical conditions on those with psychological difficulties. The National Comorbidity Survey Replication (Druss, Hwang, Petukhova, Sampson, Wang and Kessler, 2009) demonstrated that 51.8% of individuals with a chronic health condition also presented with at least one of bipolar disorder, major depression or post-traumatic stress disorder. This study demonstrated that chronic physical health conditions are more likely to be treated than psychiatric conditions (58% vs 21%); and that medical conditions become significantly more impairing for those who demonstrate comorbid psychiatric illness. These complex relationships and the deleterious impact for individuals necessitates further research to better understanding the mechanisms of action. One possibility is that an underlying mechanism impacts both physiological and psychological health. As discussed in Chapter One, previous research has linked the physiological stress-response system with psychological health. The focus of this chapter is to examine the stress-response system and establish whether measures of the stress-response system are able to predict depression, anxiety and wellbeing during adolescence.

4.2 Background literature and previous findings

The Stress Response

The stress response system is an evolutionarily driven equilibrium regulated by homeostasis, which involves the appraisal of stimuli in order to elicit adaptive biological and behavioural responses to either maintain or re-establish the equilibrium. The 'stress response' refers to the biological and behavioural responses, while stress refers to the state evoked by the stressor. The limbic system (hippocampus, amygdala and frontal cortex) comprises the interface between stressor appraisal and stress response. Physiological components of the stress response elicit functions to aid survival (e.g. increased availability of energy due to enhancement of gluconeogenesis, protein catabolism and lipolysis). An increase of respiratory

and heart rate and blood pressure allows greater availability of oxygen. Concurrently, functions that are not required for immediate survival are down regulated; for example, digestive functions, endocrine processes for growth and reproductive behaviour and immune functions. A surge of focused attention, alertness, vigilance and cognition allow for information-gathering behaviours in potentially threatening situations (see Sharpley, 2009)

Stress Response and Psychological Health and Wellbeing

This stress response mechanised by the HPA axis is thought to be a major physiological mechanism through which stress influences disease risk. It is considered that dysregulated, and particularly chronic activation of the HPA axis, causes increased wear and tear of bodily systems. As introduced in Chapter One, a growing body of evidence implicates affect with a broad variety of health outcomes (Hoyt, Craske, Mineka & Adam, 2015). Research particularly indicates a relationship between mood and cortisol; specifically, negative affect has been associated with dysregulation of daily cortisol secretions as measured via HPA axis activity. For example, Adam (2006) found that each standard deviation increase of measures of worry was associated with a 5% increase in cortisol measures. High cortisol levels have potential deleterious effects; cortisol exposure has been linked to various physiological, emotional and behavioural processes (McEwan, 1998).

Measuring HPA Axis Activity

Indicators of cortisol (the endpoint of the HPA-axis) functioning can be assessed in a variety of ways. Research has examined cortisol production across the whole day as well as patterns of changes within the day. As discussed in Chapter One, normal cortisol production follows a diurnal pattern of secretion (see Figure 4.1). As such, time of cortisol sampling is important. Cortisol levels typically are high upon waking, increase to peak levels approximately 30-45 minutes after waking, and decrease steadily throughout the day to near zero levels at bedtime (Kirschbaum and Hellhammer, 1989). The study described within this chapter will examine absolute cortisol levels at three time points (bed time, waking and peak levels) as well as measures of cortisol awakening response and hair cortisol concentration. Differences between the measures employed in this Chapter are discussed below alongside findings of previous research employing these measures.

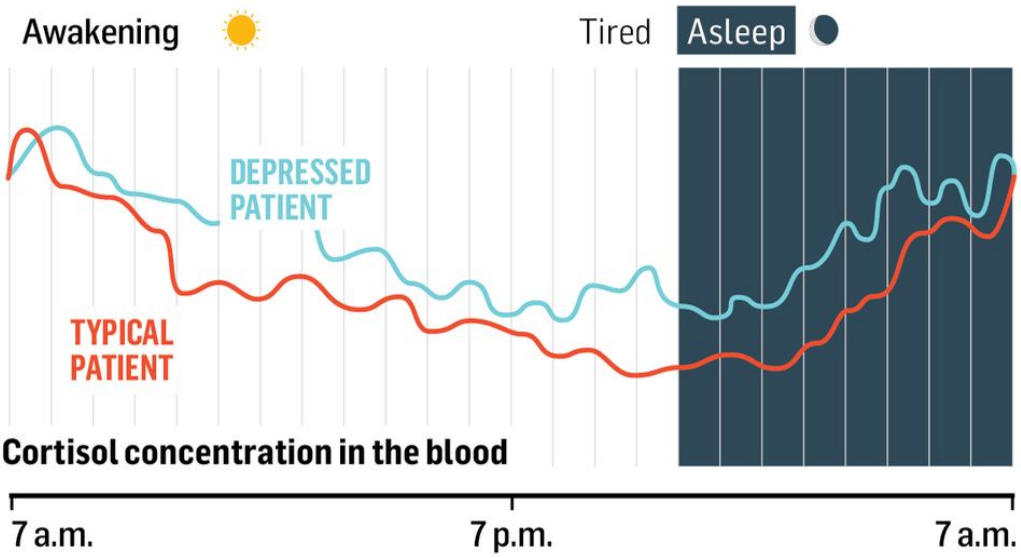


Figure 4.1: Data source: P.W. Gold and G.P. Chousos, Molecular Psychiatry, July 2002; featured in Popular Science (2015)

Temporally Variable Cortisol Measures

Absolute measures of evening and morning cortisol have been employed within research, typically via saliva samples. These measures are considered to capture levels of free circulating cortisol in plasma. Single day sampling of cortisol has been considered to estimate state rather than trait values (Adam and Kumari, 2009). Research typically employs measures of morning and evening cortisol to assess the diurnal variation. Waking cortisol measures refer to samples taken immediately after waking; sampling of peak cortisol levels tends to occur approximately 30 minutes after waking (Dedovic and Ngiam, 2015; see Figure 4.1 and 4.2); and, and bedtime (also referred to as evening) cortisol measures refer to samples taken at bed time.

As well as assessing absolute levels of cortisol, calculations of change between time points have also been examined. Measures of 'cortisol awakening response' ('CAR') has been employed within research. Typically, CAR refers to the sharp increase of cortisol secretion following waking. Consequently, CAR measures individuals' cortisol reactivity. CAR is considered to be distinct from general basal cortisol secretion as it is regulated differently than diurnal cortisol production and there is little association between the magnitude of CAR and cortisol production throughout the day (Steptoe and Serwinski, 2016). Furthermore, CAR has been proposed to be part of the waking process and therefore reflective of an anticipatory response preparing individuals for upcoming daily demands (Steptoe and Serwinski, 2016; Wilhelm, Born, Kudielka, Scholts and Wust, 2007).

Recent review has implicated CAR as a factor associated with vulnerability to depression in adult samples. Although the direction and mechanisms of the relationship between CAR and depression are complex and (so far) unclear, Dedovic and Ngiam (2015) considered CAR to be more sensitive to changes of depression than previously employed measures of the HPA axis, specifically dexamethasone suppression testing. CAR is considered important for a variety of waking processes including restoration of consciousness, alertness, hormones and the immune system.

One significant problem within this research is that CAR has been measured in different ways. Previous research has calculated CAR based on the change of absolute values across various time points or using area-under-the-curve (AuC) summary measures (see Figure 4.2). AuC measures can refer to increase from waking (AuCi) or from ground/baseline, (AuCg). Calculations based on absolute values and AuC have been used interchangeably between research groups making comparisons between studies difficult. Consensus Guidelines (Stalder et al., 2016) recommend that CAR calculations are based on area under the curve calculation, with respect to increase and ground. As such, this study has used measures of CAR based on AuC calculations rather than subtracting absolute values.

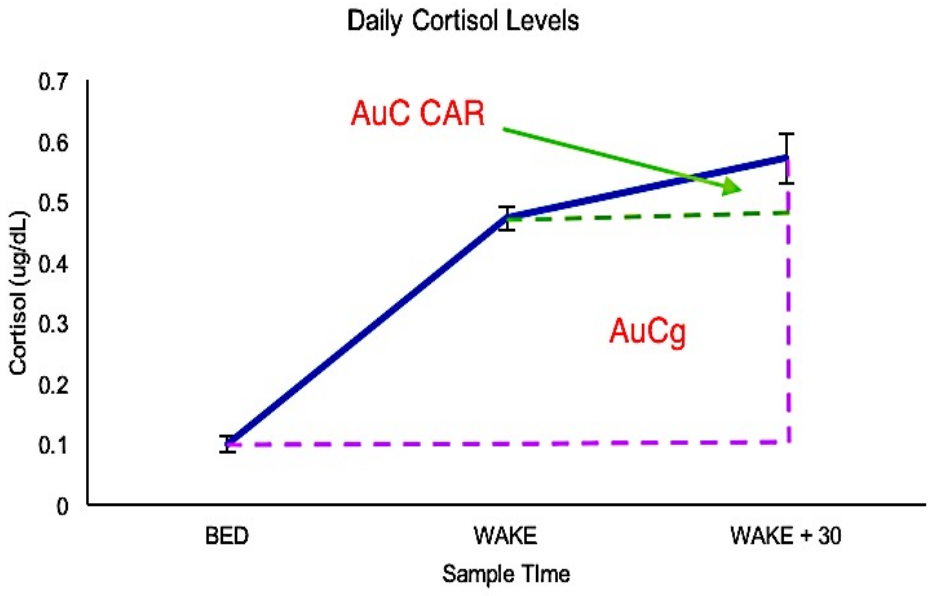


Figure 4.2: Area Under the Curve Calculations

Longer Term Cortisol Measures

The hypothesised deleterious effects of cortisol dysregulation are likely to be a consequence of prolonged dysregulation over time. It has been posited that chronic cortisol exposure results in a broad range of negative health outcomes and acceleration of disease development of both

physical and psychiatric disorders (McEwen, 2000). However, cortisol measured via saliva captures the amount of free circulating cortisol, which is highly temporally variable. Hence, analysis of this measure is not appropriate for assessing levels of cortisol exposure over a longer time period. Cortisol measured from hair samples may serve as a valid longitudinal biomarker of chronic stress (also see Chapter One). Karlen et al., (2011) demonstrated that levels of cortisol were related to experience of serious life events, perceived stress and psychological problems during three months prior to assessment in university students; and conclude that cortisol measured in hair can be a useful retrospective biomarker reflecting exposure to major life stressors. However, Cowen (2002) posited that the majority of patients with moderate depression do not hypersecrete cortisol and that further study is needed to establish differences in the stress-response system.

As this PhD study has employed analysis based upon absolute levels of cortisol concentration at three time points, AuC based measures of reactivity, and hair cortisol concentration, literature employing each types of measurement is described below.

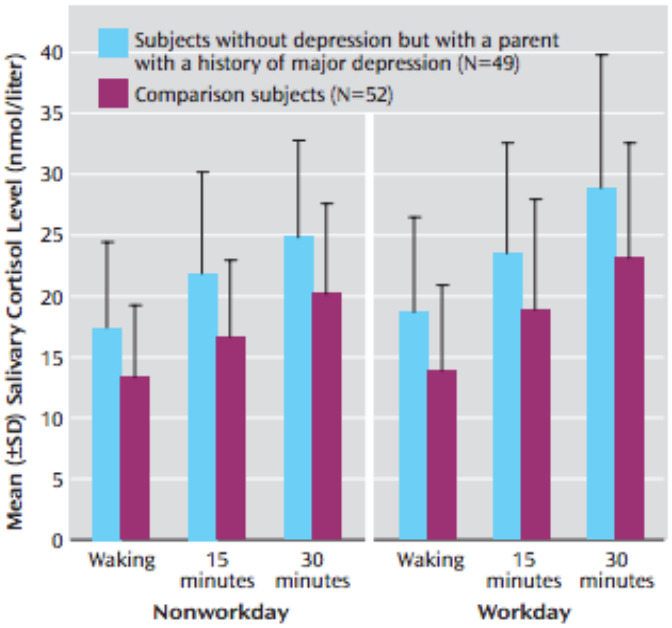
Adolescence

Within the context of adolescence, there is discrepancy between research findings. Adolescents' young age means that any deleterious costs exerted have the potential for life-long implications. Furthermore, as research has identified psychosocial and biological developmental factors as potentially involved with HPA axis dysregulation, adolescents are particularly at risk due to the significant biopsychosocial changes experienced at this age. As such, it is imperative to increase understanding of the impact of HPA axis activity in this age group. Furthermore, this developmental stage provides a unique opportunity for preventative intervention to avoid harm, mitigate risk and boost positive wellbeing.

Findings of differences in cortisol production amongst adolescents are particularly inconsistent. Consequently, Kaufman, Martin, King and Charney (2001) posited that child and adolescent depression is biochemically distinct from adult depression, specifically evidenced by cortisol secretion. They suggested that while hypersecretion of cortisol is frequently found in depressed adults, it is rarely a reported finding within children and adolescent samples. Furthermore, Kaufman et al. (2001) argued that diurnal production of cortisol in children and adolescents may be dysregulated in more subtle ways than have been seen in adults which may explain the variation of findings within this area of research (see Kaufman et al., 2001). They suggested differences may be attributed to developmental factors, stage of illness and the

heterogeneity of clinical outcome. This developmental distinction of disorder has been empirically supported by studies examining HPA axis reactivity and depression in adolescents. For example, Goodyer, Tamplin, Herbert and Altham (2000) found no association between high cortisol secretion and Mood and Feelings Questionnaire (MFQ) score, indexing depression, or stressful life events in adolescents. Nevertheless, they did find that those with high morning cortisol (levels above the 80th percentile) was predictive of diagnosis of depression two to twelve months later.

In a sample of 17-21-year-olds, Mannie, Harmer and Cowen (2007) demonstrated that never depressed individuals with a family history of depression, exhibited increased CAR compared to those with no family history (see Figure 4.3). This may indicate that increased cortisol represents a biological marker of risk to depressive illness which may have heritable underpinnings. However, Mannie et al., (2007) demonstrated that this could not be accounted for by current symptoms (based on the MFQ) or perceived stress, suggesting that elevated cortisol may constitute a broader predisposing factor rather than a marker of current disorder.



[a] Participants took samples immediately after awakening and then twice more at 15-minute intervals. Salivary cortisol levels were significantly greater on workdays than on nonworkdays (F=10.13, df= 1, 99, p=0.002) and significantly greater in the FH+ group on both days (F=15.46, df=1, 99, p<0.001).

Figure 4.3: Mannie et al. (2007): Comparison of morning salivary cortisol levels in participants at high familial risk for depression versus controls

A ten-year longitudinal study examined the premorbid cortisol secretions in adolescents (Mathew, Coplan, Goetz, Feder, Greenwald, Dahl, Ryan, Mann and Weissmann, 2003). No

differences in cortisol concentrations between individuals who developed depression and those who remained healthy were revealed. However, this study found that those who had experienced depression and attempted suicide over the follow up period (n=13) had significantly higher overall cortisol levels (measured via blood samples every 20 minutes over 24-hours) when compared with those who had developed depression but did not attempt suicide (n=23); those who had attempted suicide at the time of baseline assessment (n=20); and, those who had no history of depression (n=21). Interestingly, Matthew et al., (2003) demonstrated that adolescents who were at risk of suicide attempts demonstrated elevations of cortisol concentration prior to sleep onset, when cortisol is typically at its lowest. Although this study did not directly implicate cortisol in depressive onset, and is likely underpowered, its results have potentially important clinical relevance in that individuals with suicidal attempts and depression may be distinguished by biological markers.

Similarly, in a sample of 230 adolescents (aged 16-18), higher baseline CAR was associated with an increased risk of developing depression within a 1-year follow-up period (Adam, Doane, Zinbarg, Mineka, Craske and Griffith, 2010, see Figure 4.4). Nevertheless, unlike Goodyer et al., (2000) which found the use of absolute cortisol values were not predictive of subsequent disorder, Adam et al., (2010) demonstrated that absolute values were associated with an increased risk of developing depression (Odds Ratio=3.0, p=.04). Furthermore, this association remained significant after controlling for baseline depression score and results indicated this association was not accounted for by stressful life events.

Similar to Chapter Two and Three, the empirical study presented in this chapter seeks to examine the full spectrum of mood states rather than focusing exclusively on clinically depressed adolescents, including a measure of wellbeing in addition to psychopathological symptoms (see Chapter One). Limited research has examined wellbeing in relation to HPA axis functioning using measures of cortisol, conclusions are difficult to draw due to variation of sample age, disorder status and protocols of cortisol measurement.

One aim of this thesis is to explore the role of wellbeing in relation to risk factors for disorders, as wellbeing may constitute a protective factor against disorders. Only one study has examined wellbeing in relation to cortisol production within an adolescent sample (Rickard, Chin and Vella-Brodrick, 2016). Rickard et al., (2016) demonstrated significant partial correlations between CAR and multiple measures of wellbeing when controlling for stressful life events. When employing a hierarchical multiple regression approach, CAR significantly improved the

prediction of mental wellbeing by 19% more than that accounted for by life events alone. This indicates that measures of CAR may be useful markers of wellbeing and positive functioning in addition to distress within adolescents.

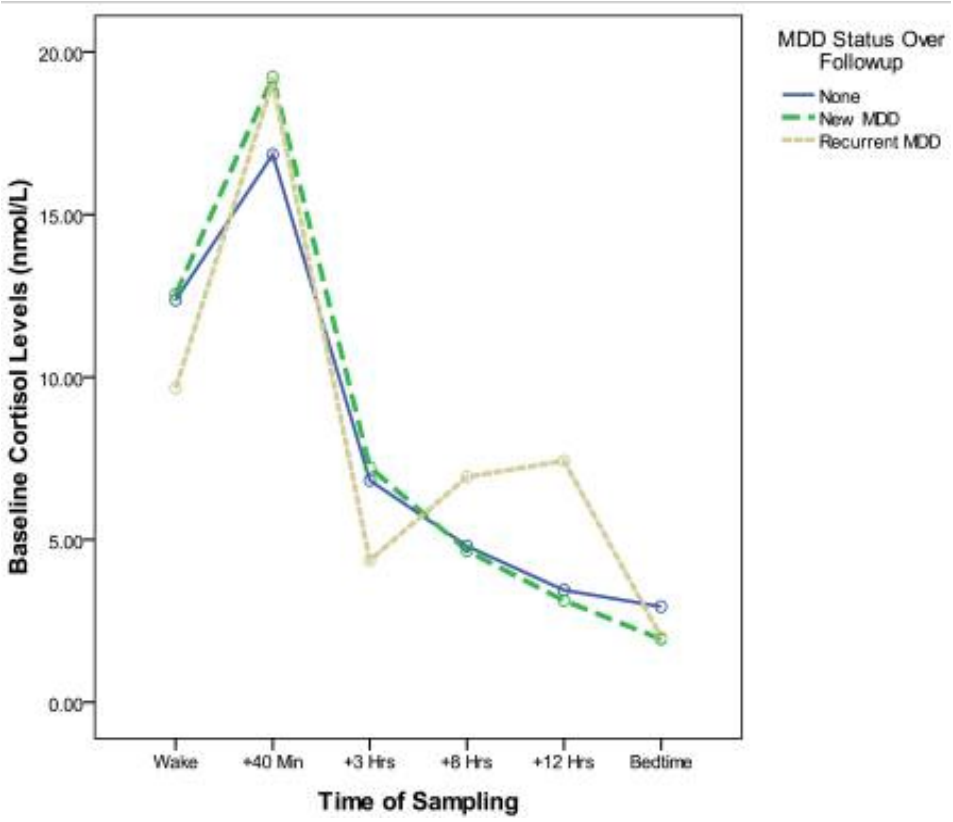


Figure 4.4: Adam et al., (2010) Baseline diurnal cortisol profiles and depression status over the following year

Unexpectedly, Rickard et al. (2016) failed to demonstrate that CAR was significantly predictive or correlated to anxiety, which was measured alongside wellbeing and negative affect. This is somewhat surprising due to the strong associations between anxiety and depression, as well as the hypothesised relationship between anxiety and the HPA-axis implicated in stress reactivity and 'flight or fight' responses. A prospective longitudinal study demonstrated that CAR measured at age 17 was a significant prospective predictor of the onset of anxiety disorders six years later (Hazzard Ratio=2.20, 95% *CI*= [1.05-4.62], *p*=0.04; Adam, Vrshek-Schallhorn, Kendall, Mineka, Zinbag and Craske, 2014). However, waking cortisol levels were not significant. Further investigation identified that this significance was driven by the strong and significant relationship between CAR and social anxiety. When predicting social anxiety and all other anxiety disorders separately, predictions of other anxiety disorders were no longer significant.

The adolescent literature exploring the HPA axis is particularly limited, due to methodological variations, sample variations and the scarcity of studies exploring wellbeing. Previous research potentially has identified reduced CAR (a flattened slope) as well as higher basal cortisol levels in risk psychological disorders. Understanding factors contributing to wellbeing may provide avenues for early intervention or prevention treatment and as such are worth of further investigation, particularly within adolescent samples. Furthermore, identifying a biological risk factor could aid in the detection of risk and disorder, thereby facilitating treatment.

The Impact of Additional Stressors

Environmental, personality and psychosocial stressors are also associated with the risk for psychological illnesses. For example, a meta-analysis (Burke, Davis, Otte and Mohr, 2005) demonstrated that during stress induction and recovery, individuals with depression differ from healthy controls in response to stress. Specifically, depressed individuals were characterised by blunted stress reactivity and impaired recovery. This pattern of 'flattened' cortisol activity has also been found in adolescents experiencing their first episode of depression who also had experienced a stressful life event compared with non-depressed controls (Mazurka, Wynne-Edwards and Harkness, 2016). Furthermore, HPA axis patterns were able to distinguish the first episode and recurrent episodes of depression in relation to stressful life events, suggesting that the HPA axis response may be a neurobiological mechanism mediating the effects of stressful life events and depression (Mazurka et al., 2016).

As neuroticism is a known personality risk factor for depression in adults (Weissman, 2016; Navardy et al., 2017), it is therefore important to explore whether relationships exist between neuroticism and the HPA-axis functioning. Mannie et al., (2007) reported high correlations of state measures of mood with trait measures of personality factor neuroticism and between cortisol levels and neuroticism, and consequently reported difficulty in differentiating the relationship of these two components with cortisol. Chan et al (2007) however did not find differences in cortisol level between never-depressed adolescents with high vs. low level of neuroticism. Similarly, Windle (1994) found only a weak relationship between baseline cortisol and depression, neuroticism and anxiety. This may be due to temporal factors; the disturbance of the HPA axis may develop over time. High levels of cortisol may be first induced by constant feelings of being stressed and then result in desensitisation of receptors and lower production of cortisol. The same may be true to a lesser degree in non-clinical samples with subclinical dysphoria or depression.

In another study with an adolescent sample, Hauner et al., (2008) employed a three-level multilevel analysis to model the diurnal shape of cortisol rhythm and explore associations with neuroticism. This study found no significant main effects of neuroticism on diurnal slope, CAR or waking cortisol values. However, it did uncover a gender by neuroticism interaction, driven by males with higher neuroticism demonstrating a significantly flatter diurnal cortisol slope. This study argued that flatter slopes may be present only in men due to stress exerting a greater physiological response in men compared to women. This is in line with previous research that found males responding to laboratory stressors with higher cortisol responses than females (Kudielka and Kirschbaum, 2005 and Traustadottir et al., 2003, both in Hauner et al., 2008).

Emotion and Stress Response

Emotional reactivity and regulation may also play a role in determining the extent to which an individual is affected by exposure to stressful life events. Emotional reactivity refers to the extent to which individuals experience emotions; prevailing theory categorises this into arousal intensity, sensitivity and persistence of emotions. Measures of emotional reactivity have been related to self-reported stress and coping factors (e.g. Shapero, Abramson and Alloy, 2016). This is important as coping and stress have also been frequently aligned with HPA axis functioning and psychological disorders. It is possible that emotional reactivity triggers HPA axis activity, exacerbating health outcomes. In this sense, it may be that the physiological stress-response system reflects that of the internal psychological stress system: emotional reactivity and regulation.

Emotion regulation comprises an internal cognitive mechanism of responding to emotions. Reappraisal is considered an adaptive strategy whereby negative experiences are reframed in order to reduce their negative emotional impact. Conversely, suppression is considered a maladaptive strategy, whereby individuals attempt to suppress and avoid experiencing emotions. Empirical evidence indicates that suppression is associated with an increase of negative emotions and psychological distress. As emotional regulation is an internal mechanism controlling emotions, its associated psychological distress may exert physiological effects via the HPA axis and it may be that emotional regulation exerts or can mitigate physiological effects. The stress response system monitors and reflexively adapts physiological responses to stressors, not unlike the process of emotional regulation.

McLaughlin and Hatzenbuehler (2009) examined the role of emotional dysregulation in linking experiencing stressful events and internalising symptoms in adolescents. Mediation analysis revealed a significant indirect effect of stress on depressive and anxious symptoms through emotion dysregulation (z=5.05, p<.001), in McLaughlin and Hatzenbuehler's (2009) longitudinal study of 1,567 adolescents. In this study, the mediation results were stronger for anxiety than depression, potentially indicating greater relevance of this pathway for the understanding of anxiety. However, no studies have examined physiological, rather than subjective experiences, of stress and relationships with emotional and psychological variables.

4.3 Aims of the Present Study

This study aimed to investigate whether free circulating and longitudinal measures of cortisol are predictive of depression, anxiety and wellbeing, independent of neuroticism, within the general population of adolescent participants. Due to the variation of findings, directionality of the relationship between cortisol and psychological measures has not been hypothesised. Additionally, this study aimed to examine the relationship between measures of cortisol (via hair and saliva) and stressful life events, emotional reactivity and emotional regulation, which have been robustly linked with disorder onset and maintenance across development.

This study recruited participants from local schools. As discussed in the previous chapter, this approach has been adopted in order to overcome some of the problems and potential biases that occur when recruiting from clinical settings (see Chapter One). In keeping with understanding mental health as a spectrum rather than as distinct categories, recruiting from clinical settings would constrain the sample to those who are help-seeking, receiving help and exhibiting clinically significant symptoms (see Chapter One).

4.4 Methodology

Participants

In line with the aims of this thesis, this study sought to recruit a community sample of adolescents aged 13-18 (mean =15.65; SD=1.67). A total of 131 participants were recruited from local Scottish secondary schools. Final sample size for each measure is reported below.

Participants were considered to be eligible for inclusion if they were between 13-18 years of age and self-identified to understand and speak English fluently. Similar to Study 1, all adolescents regardless of mood symptomology were considered eligible. Individuals were required to provide informed consent and parental consent was obtained from participants under the age of 16. This study had no other exclusion criteria.

Sample Size

Previous research examining factors similar to this study, namely emotional regulation and cortisol in relation to mood, within adolescent populations, reported medium effect sizes. A power calculation using a medium effect and five predictor variables in a regression design was conducted using G*power, this indicated that a sample of 92 would be necessary (f^2=0.15, α error probability=0.05, 1-β error probability=0.80).

Measures

Demographic Information

A short form was used to gather demographic information including gender, age, ethnicity and personal and family history of mental wellbeing. Individuals were asked about any medications that they might take and approximations of height and weight, due to the influence of these factors on cortisol production.

Pubertal Timing

The Pubertal Development Scale (Petersen, Crockett, Richards and Boxer, 1987)

To account for individual differences in development, which potentially influences cortisol production, a brief self-report measure was used to assess developmental characteristics, as is common practice in similar studies within this age group. Often pubertal status is assessed by

physical examination by physicians. However, this was deemed less feasible and appropriate for this sample and study design. This self-report measure of puberty has previously demonstrated a high correlation with physician ratings of puberty indicating the validity of this measure (Petersen et al., 1987). Participants were asked to respond to five questions, specific to their self-identified gender, relating to physical development on a four-point scale (1=no development; 2= beginning development; 3=additional development; and 4=development completed). Females were asked to indicate their physical development in relation to body hair, breast change, skin change, growth spurt and menarche. Menarche was coded dichotomously with 1 indicating pre-menarcheal and 4 indicating post-menarcheal status. Boys were asked to indicate their physical development in relation to body hair, voice change, skin change, growth spurt and facial hair. Internal consistency assessment indicated that α =.82 for females and α =.68 for males, in this sample.

Psychological Measures

Each of the following measures have been deemed to be age appropriate and have previously demonstrated satisfactory psychometric properties. See Table 4.1 for means and standard deviations within this sample.

Mood and Feelings Questionnaire-Child Self-Report (MFQ; Angold et al. 1995)

This is a well-established 33-item self-report questionnaire assessing symptoms of depression based on DSM criteria. Participants were presented with a series of descriptive phrases relating to how they have been feeling or acting recently. Participants are required to respond to each statement by selecting 'not true', 'sometimes', or 'not true'. Daviss et al. (2006) found it to have excellent internal consistency (α =.95), which was replicated in this study, and to be successful in discriminating between depression and other mood disorders.

BBC Wellbeing Scale (Kinderman, Schwannauer, Pontin and Tai, 2011)

This 24-item questionnaire was used to assess three key domains of wellbeing (physical health, psychological health and relationships). This measure focuses on positive features of mood to supplement our measure of psychopathology. Participants were asked to respond on a 4-point Likert scale. The internal consistency of this scale has been previously reported with Cronbach's alpha .94 (Kinderman et al. 2011) and this study demonstrated alpha=.95. This scale has been validated in a sample of 1,932 participants, of whom 228 were school-aged

adolescents. In the present study, this scale demonstrated excellent internal consistency (α=.92).

Spence Children's Anxiety Scale (SCAS; Spence, 1997)

The SCAS is a 44-item self-report questionnaire assessing six subscales of anxiety: generalised anxiety, panic/agoraphobia, social phobia, separation anxiety, obsessive-compulsive disorder, and physical injury fears. Participants are asked to rate the degree to which they experience each symptom on a four-point scale. This questionnaire has demonstrated good psychometric properties in both clinical and non-clinical populations of adolescents, (α=.93) and correlations between other measures of anxiety were high (r=.79; Muris, Merckelbach, Ollendick, King and Bogie, 2002). In this study, this scale demonstrated excellent internal consistency (α=.95).

Personality

Eysenck Personality Questionnaire- Neuroticism (EPQ-N; Eysenck, Eysenck and Barrett, 1985)

Neuroticism was evaluated using the shortened form of the neuroticism scale (12 items), from the Eysenck Personality Questionnaire. This is a well-known and widely used measure of personality traits and has been used with adolescent samples previously (e.g. Kuyken, Watkins, Holden and Cook, 2006). In this study, this scale demonstrated good internal consistency (α=.83).

Emotional Reactivity and Regulation

Emotional Reactivity Scale (ERS, Nock, Wedig, Holmberg and Hooley, 2008)

The ERS is a 21-item self-report measure assessing individuals' experience of emotion reactivity. This scale was developed for use within adolescent samples (mean age=17.1, S.D.=1.88, range 12-19). Each statement is responded to on a four-point scale (0= not at all like me, and 4= completely like me and comprises of three subscales assessing sensitivity, arousal and persistence. The ERS has demonstrated excellent internal consistency (α=.94) and construct validity in previous research (Nock et al., 2008), in this study this scale demonstrated excellent internal consistency (α=.95).

Emotion Regulation Questionnaire for Children and Adolescents (ERQ-CA; Gullone and Taffe, 2011)

In keeping with Gross' model of emotional regulation, which separates emotional regulation into the strategies of either cognitive reappraisal or expressive suppression (Gross and John, 2004), the ERQ-CA was employed here. The ERQ-CA is a 10-item self-report measure of individuals' tendencies to employ reappraisal (six items) and suppression (four items) to regulate emotion. It is based on the adult scale developed by Gross and John (2003). Each item is measured on a five-point Likert scale with one indicating 'strongly disagree' and five indicating 'strongly agree'. This study demonstrated acceptable-good internal consistency (α=.76-85), an improvement from previous reports (Gullone and Taffe, 2011, who demonstrated adequate internal consistency of the scale (α=.69-79).

Cortisol Measures

Hair

Longer term cortisol exposure can be assessed through measurements of cortisol concentration within hair samples. Hair stores cortisol following exposure to blood flow within the skin and grows at approximately once centimetre per month. Therefore, hair samples can provide a retrospective measure of longer-term exposure (Stalder and Kirschbaum, 2012). Hair samples, approximately 1cm wide, were cut close to the scalp in the posterior vortex area of the head, as is standard practice due to this area demonstrating the lowest coefficient of variation (Suave, Koren, Walsh, Tokmakejian and Van Uum, 2007). Hair samples were tied with an elastic band at the end closest to the scalp, wrapped in tin foil, labelled and stored in a freezer at -20^{0}C within Tommy's Centre for Maternal and Fetal Health Research (within the Queen's Medical Research Institute, 47 Little France Crescent, Edinburgh, EH16 4TJ) until all samples were collected and ready for analysis. Analysis of hair has shown to be unaffected by dyes, chemical straightening or perming (e.g. Suave, Koren, Walsh, Tokmakejian and Van Uum, 2007).

Saliva

In a commonly employed method and utilising 'Sarstedt Salivette' collection kits, saliva samples were collected from participants to measure evening and early morning cortisol response. Following the manufacturer's instructions, participants were asked to chew on a cotton swab for 60 seconds immediately prior to bed and then spit the cotton into the plastic collection tube and seal it. This procedure was repeated immediately upon waking and 30 minutes following waking. Participants were also asked to fill in the date and time of sample collection on the label of the tube. Participants were asked to refrain from eating, drinking or brushing teeth 30 minutes prior to taking the sample.

Stressful life events

Life Events Rating Scale (adopted from Goodyer, Herbertm Tamlin, Secher and Pearson, 1997)

This measure assessed a variety of events that may be considered to be stressful over the past 18 months, for example, moving school, changes in family arrangements and illness in relatives or close friends. Participants responded to 12 questions giving yes/no response as well as describing the event (if applicable). Participants were also asked to rate each experience on a 5-point scale (1=very pleasant/happy, 5=very unpleasant/sad/painful), if participants responded to the scale with either a four or five, they were asked if this has affected them for more than two weeks, requiring a yes or no response. Participants are asked for approximate dates of experiences. A 13th question, with the same question format, asked if there are any other important events that have happened prior to the previous 18 months. In line with previous research (Goodyer et al., 1997), when scoring this scale, a stressful life event was defined as any event that happened within the past 18 months that was rated as either 4 or 5 and reported to impact them for more than two weeks. The number of stressful life events was totalled for each participant.

Procedure

Following ethical approval from the University of Edinburgh, School of Health in Social Science Ethics Board and relevant Council Education authorities, schools within Edinburgh were contacted to provide information about the study and asked if they would be interested in facilitating participation. A meeting was arranged to further discuss participation with schools that indicated interest.

School pupils were invited to an information event where they were provided with information packs which included Participant Information Sheets, Parental Information Sheets, Consent Forms, Parental Consent Forms and Assent Forms. If they wished to take part, informed consent was obtained from individuals over the age of 16 and parental consent (and individual assent) in the case of individuals under the age of 16. At this stage, and throughout, all individuals had the opportunity to ask any questions.

After consent was obtained a suitable time was arranged to return to the schools to provide individuals with the equipment and specific participation instructions. All questionnaires were administered online, hosted securely via Bristol Online Survey. Participants were required to insert their participant number in order to access the questionnaire, and to allow for the matching of questionnaires with other components. The online survey was estimated to take approximately 20-30 minutes and comprised of: demographic and basic information (see measures) and the MFQ, SCAS, BBC, EPQ-N, ERS, ERQ-CA and Stressful Life Events questionnaires.

A hair sample was also taken at this meeting. The procedure for the collection of saliva samples was also explained to participants at this meeting, and written instructions for all components of participation were provided. The days on which the saliva samples would be deposited was agreed with participants.

Participants had one working week to complete the online questionnaires. The researcher collected saliva samples from participants on the agreed day. At this point participants were debriefed via a debrief form and received a £10 voucher for their participation. If online questionnaires had not been completed at this stage, participants were prompted and reminded of the online link address.

Processing of Biological Samples

Saliva samples were collected, processed and frozen on the same day as the morning samples were deposited, other than in cases where participants forgot to return samples on time. In these cases, samples were collected and processed at the soonest available opportunity. Saliva was extracted from the cotton swabs, labelled (by participant ID and date) and stored securely in a freezer within Tommy's Centre for Maternal and Fetal Health Research (within the Queen's Medical Research Institute, 47 Little France Crescent, Edinburgh, EH16 4TJ), at -20^{o}C along with hair samples until analysis after all samples had been collected.

Saliva samples were analysed using Cortisol ELISA kits as per manufacturer's instructions by researchers within the Queen's Medical Research Institute (see Salimetrics Salivary Cortisol Enzyme Immunoassay Kit, Salimetrics.com, 2016, for full details). Cortisol concentration of hair samples was analysed on site by researchers at Tommy's Centre for Maternal and Fetal Health Research. A three-centimetre section of hair was cut from the scalp end of the hair sample and weighed to ensure all samples weighed over 25mg. Hair cortisol concentration

analysis has been described by Binz, Braun, Baumgartner and Kraemer (2016). Briefly, this procedure involves a two-step washing with water then acetone procedure, 3cm hair samples were cut into smaller segments. Samples were spiked with the internal standard and incubated overnight. Cortisol was assessed via a liquid chromatography-tandem mass spectrometry (LC-MS/MS) system. Extreme high and low values were reanalysed to ensure correctness.

4.5 Statistical Analysis

Prior to analyses, distributions skewness and kurtosis of raw data were examined. Extreme outliers that constituted improbable values were considered errors (for example, one participant reported a BMI of 155) and were subsequently removed; this was the case for five data points (3 BMI values, one salivary cortisol concentration value and one hair cortisol concentration value).

Salivary and hair cortisol measures were found to fit lognormal distributions, and as conceptually values cannot be negative, a log transformation was applied (Adam and Kumari, 2009). Following which, data were judged to fit the normal distribution. All subsequent analyses were conducted with transformed variables. Graphs displayed below are based on untransformed data.

Puberty was found to not fit a normal distribution (see Figure 4.3) and that this variable ought to be employed in a categorical fashion, but due to the limitations of the sample size this could not be done. Despite this, puberty has been entered in regression models despite this. Consequently, caution should be taken when interpreting results.

Similarly, although medication use has been measured it has not been included in analysis due to its categorical nature. Collection of this variable was considered important in the event that individual cortisol concentration fell outwith the expected range.

Following the exclusion of data points based on the above criteria, and missing data 58 participants had no missing cortisol data. Four participants with complete cortisol data failed to complete psychological measures. The sample size for each measure can be seen in Tables 4.1 and 4.2.

Saliva samples were excluded from analysis if time information had not been provided. Furthermore, Wake + 30 samples were excluded if the time of the deposited sample outwith a 15-minute window of the 30 minutes projected from the wake sample. Previous research has utilised sampling up to 45 minutes from waking to examine CAR (Adam and Kumari, 2009)

In line with previous research (e.g. Golden, Sanchez et al., 2014), values were calculated, using R Studio (Version 1.1.383), via an area under the curve calculation using the trapezoidal rule (e.g. Pruessner et al., 2003) with respect to ground (AuCg) and increase (AuC_i), to represent total cortisol production and CAR (see Figure 4.1). Hair cortisol concentration was analysed by in-house researchers of the Queens Medical Research Institute. Values of cortisol concentration are reported in (pg/mg). Three cm segments of hair from the end closest to the scalp were cut (five samples were not long enough and were 2 or 2.5 cm in length). Samples were weighed to ensure they were 7.5mg, four samples weighed less than 7.5 mg (3.0, 3.4, 5.6 and 6.7 mg). These samples were not removed as their cortisol concentrations were within the expected ranges. One sample was excluded due to extremely high cortisol concentration (107.64 pg/mg). Adam and Kumari (2006) discuss the variation of cortisol measures and state that approximately 1% of cortisol measures are found to be substantially above the mean although they are not clear as to what such high measures represent. They conclude that there is not an established practice in terms of windsorising versus excluding outliers within this context. As such, this extreme outlier was removed to minimise its impact on the data.

The following measures were examined -

1. Absolute cortisol levels:
    i. bed time cortisol concentration (bed),
    ii. waking cortisol concentration (wake),
    iii. peak cortisol concentration (wake+30),
2. Change values reflecting reactivity:
    i. AuC calculation with respect to ground/baseline (AuCg),
    ii. AuC calculation with respect to increase (AuCi; represents CAR)
3. Longitudinal hair cortisol concentration (hair).

To examine the relationship between cortisol and depression, anxiety and wellbeing, forced entry hierarchical regressions have been employed to predict depression, anxiety and wellbeing in turn, as in previous chapters. Firstly, regression models used saliva based (log transformed) bed, wake and wake+30 cortisol concentration values. Secondly, regression

models used saliva based AuC calculations (AuC_g and AuC_i, representing individuals' cortisol reactivity and; longitudinal cortisol measures (hair sample) and theses were also included in these models.

For both sets of regression models, depression, anxiety and wellbeing were predicted in turn. Initial blocks included control variables, age, gender, puberty and BMI. Neuroticism was included in the second block, to account for variation due to personality risk. The third and final block included cortisol variables. Changes in r^2 and beta coefficients have been reported and considered throughout.

Due to the limited power to include further variables in regression analysis, bootstrapped Pearson's bivariate correlation examined any relationships between cortisol measures with stressful life events, emotional reactivity and regulation, while controlling for age, gender, puberty and BMI. Results are based on 1000 bootstrapped samples.

4.6 Results

Initially, descriptive statistics and age and gender differences are reported (see Table 4.1 and Table 4.2).

Table 4.1: Descriptive Statistics of Questionnaire Measures

Measure	N	Mean	S.D.
Pubertal Development	104	16.68	2.78
MFQ	104	17.32	13.71
SCAS	104	30.29	19.67
BBC	104	66.36	12.32
Neuroticism	104	6.73	3.47
ERS	104	45.89	16.10
ERQ: Reappraisal	104	18.43	4.42
ERQ: Suppression	104	12.12	3.43
Stressful Life Events	104	2.81	2.23

Table 4.2: Descriptive Statistics of Cortisol Measures

Measure (units)	N	Minimum	Maximum	Mean	S.D.
Bed **(ug/dL)**	89	BTD	0.86	0.10	0.13
Wake **(ug/dL)**	100	0.14	1.04	0.47	0.19
Peak **(ug/dL)**	93	0.06	1.06	0.53	0.21
AuC_g **(ug/dL)**	82	0.39	1.35	0.79	0.26
AuC_i **(ug/dL)**	93	0.23	0.93	0.50	0.17
Hair **(pg/mg)**	102	0.67	42.06	10.32	8.51
BDT= below threshold of detection					

Age, Gender, Puberty and BMI

The mean age of participants was 15.65 (SD=1.67), 60.9% of the sample was female and participants' mean BMI was 20.38 (SD=3.04). The scoring of the pubertal development scale resulted in scores between 5 and 20 (see Figure 4.5 and Table 4.1), mean puberty score for the sample was 16.68 (SD=2.78).

Age and Gender Differences

Figures 4.6 and 4.7 depict cortisol concentration for males and females. T-tests did not indicate significant gender differences in cortisol concentration, but a significant gender difference in pubertal development; females demonstrated greater pubertal development (mean=17.06, SD=2.82) compared to males (mean=15.35, SD=2.21; t(102)=2.69, p=0.008). Gender differences were also indicated in measures of anxiety (SCAS) where females demonstrated higher levels of anxiety (mean = 32.47, SD=20.80) than males (mean=22.61, SD=12.66; t(102)=2.16, p=0.03). No other measures demonstrated significant gender differences.

Bivariate Pearson's correlation was employed to assess the effect of age. Age was significantly correlated with puberty (r^2=0.23 p<0.001), BMI (r^2=0.04, p<0.04); stressful life events (r^2=0.04, p=0.04) and negatively correlated with hair cortisol levels (r^2=0.12, p<0.001). This indicates that increasing age was correlated with increased pubertal development, higher BMI,

more stressful life events and reduced cortisol as measured via hair samples.

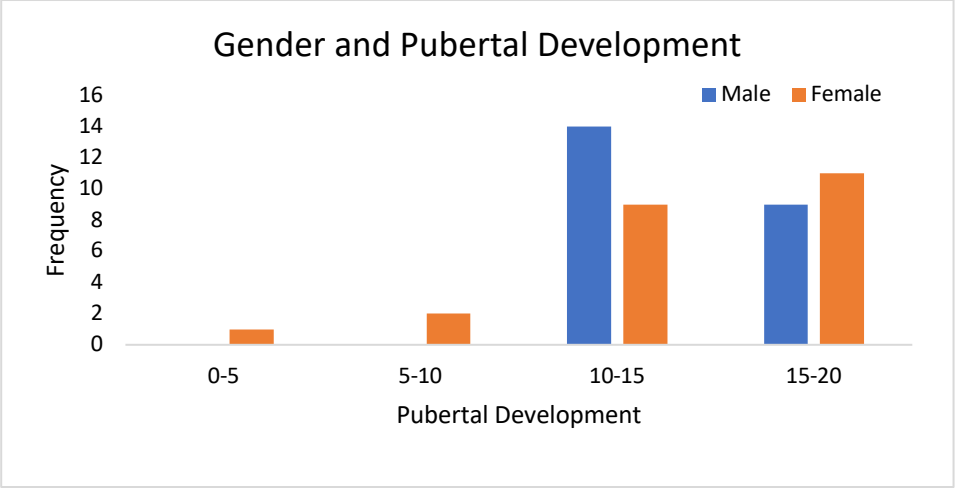


Figure 4.5: Distribution of Pubertal Development within Gender

To confirm that salivary cortisol data followed the expected diurnal trend, mean cortisol concentrations were plotted at each time point (see Figure 4.4). This demonstrates, as expected, lowest levels of cortisol were observed in the evening, spiking at waking with a further increase 30 minutes after waking. See Table 4.2 for descriptive statistics of salivary and hair cortisol values

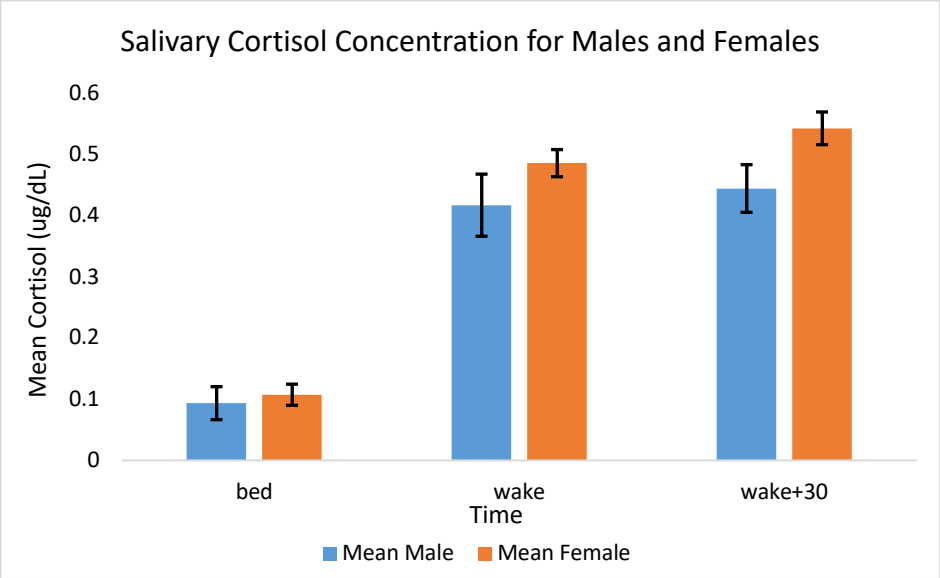


Figure 4.6: Mean Salivary Cortisol Concentration

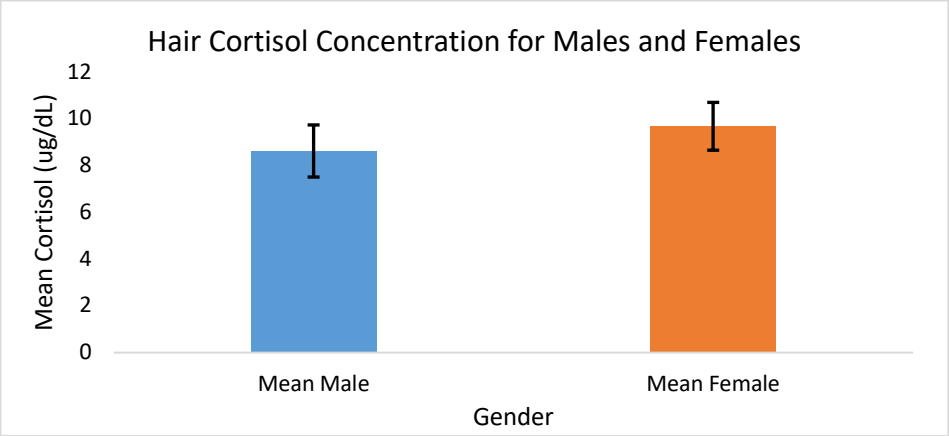


Figure 4.7: Male and Female Hair Cortisol Concentration

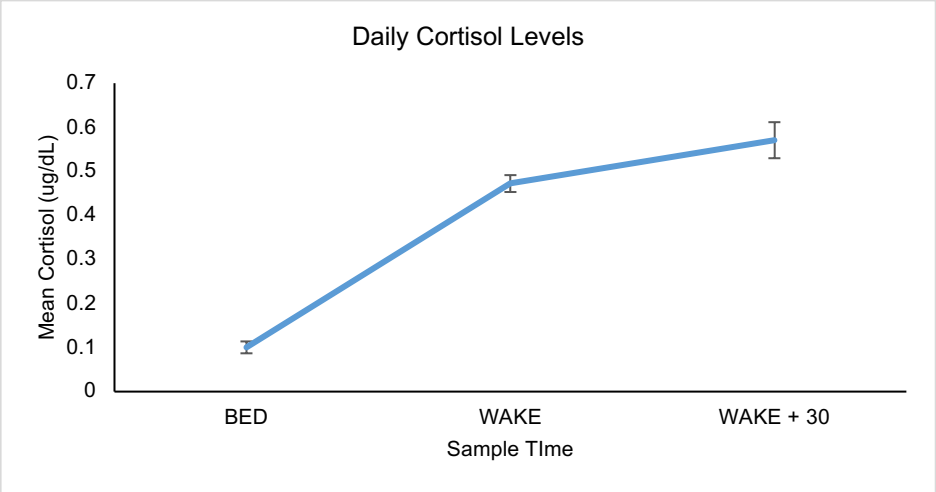


Figure 4.8: Mean Daily Cortisol Levels

Predicting Depression, Anxiety and Wellbeing With AuC Cortisol Calculations (Inter-Individual Reactivity and Longitudinal Hair Cortisol Concentration

Table 4.3 displays the results of forced entry hierarchical multiple regression modelling, using AuC calculations of cortisol reactivity (AuC_g and AuC_i) and the longitudinal measure hair cortisol concentration. Control variables entered in the first block (age, gender, puberty and BMI) did not result in significant regression models in predictions of MFQ, SCAS or BBC. The second step, involving the addition of neuroticism demonstrated a significant increase in r^2 for each of MFQ, SCAS and BBC, with significant beta coefficients in each case (see Table 4.3). The third step, entering of AuCg, AuCi and hair cortisol concentration failed to

demonstrate a significant increase in r^2 in models of MFQ, SCAS and BBC, no standardised beta coefficients of cortisol variables were significant in these models.

Table 4.3: Hierarchical Regression Predicting Depression, Anxiety and Wellbeing using Cortisol Reactivity and Longitudinal Cortisol Measures (n=73)

Model	**Predictor**	**r**	**r^2**	**Adj r^2**	**F-value (df)**	**Unstandardised Coefficients**		**β**	**t-value**	**Δr^2**	**ΔF (df)**
						B	**SE B**				
MFQ		0.26	0.07	0.02	1.27(4,68)					0.07	1.27(4,68)
Step 1	Constant					16.79	21.15		0.79		
	Age					-1.89	1.35	-0.20	-1.40		
	Gender					5.06	4.95	0.13	1.02		
	Puberty					0.63	0.84	0.11	0.75		
	BMI					0.76	0.65	0.14	1.17		
Step 2		0.70	0.48	0.44	12.37(5,67)**					0.41	52.90(1,67)**
	Constant					9.92	15.96		0.62		
	Age					-1.37	1.02	-0.14	-1.34		
	Gender					4.28	3.73	0.11	1.15		
	Puberty					0.06	0.64	0.01	0.09		
	BMI					0.31	0.50	0.06	0.63		
	Neuroticism					2.71	0.37	0.65	7.27**		
Step 3		0.70	0.49	0.42	7.53(8,64)**					0.01	0.20(3,64)
	Constant					13.97	17.53		0.80		
	Age					-1.31	1.08	-0.14	-1.22		
	Gender					5.30	4.05	0.13	1.31		
	Puberty					0.04	0.66	0.01	0.06		
	BMI					0.24	0.52	0.04	0.47		
	Neuroticism					2.71	0.38	0.65	7.13**		
	AuCg					-2.32	15.12	-0.04	-0.15		
	AuCi					-3.37	23.25	-0.04	-0.15		
	Hair Cortisol					-0.18	0.88	-0.02	-0.20		
SCAS		0.26	0.07	0.01	1.21 (4,68)					0.07	1.21 (4,68)
Step 1	Constant					9.64	29.42		0.33		
	Age					-1.94	1.88	-0.15	-1.03		
	Gender					4.49	6.88	0.08	0.65		
	Puberty					1.67	1.17	0.21	1.43		
	BMI					0.95	0.91	0.13	1.05		
Step 2		0.77	0.60	0.57	20.05 (5,67)**					0.53	89.14 (1,67)**
	Constant					-1.23	19.45		-0.06		
	Age					-1.11	1.25	-0.08	-0.89		
	Gender					3.26	4.54	0.06	0.72		
	Puberty					0.77	0.78	0.10	0.99		
	BMI					0.24	0.60	0.03	0.39		
	Neuroticism					4.29	0.45	0.75	9.44**		

Step 3		0.78	0.61	0.56	12.60 (8,64)**					0.012	0.68(3,64)
	Constant					2.18	21.14		0.10		
	Age					-1.00	1.30	-0.08	-0.77		
	Gender					4.30	4.89	0.08	0.88		
	Puberty					0.88	0.80	0.11	1.11		
	BMI					0.06	0.62	0.01	0.10		
	Neuroticism					4.29	0.46	0.74	9.36**		
	AuCg					-0.11	18.24	0.00	-0.01		
	AuCi					-11.57	28.04	-0.09	-0.41		
	Hair Cortisol					0.66	1.07	0.05	0.62		
BBC		0.31	0.10	0.04	1.78(4,68)					0.10	1.78(4,68)
Step 1	Constant					98.13	17.52		5.60**		
	Age					0.28	1.12	0.04	0.25		
	Gender					1.55	4.10	0.05	0.38		
	Puberty					-0.99	0.69	-0.20	-1.43		
	BMI					-1.04	0.54	-0.23	-1.93		
Step 2		0.63	0.40	0.35	8.86(5,67)**					0.30	33.75(1,67)**
	Constant					103.09	14.42		7.15**		
	Age					-0.10	0.92	-0.01	-0.11		
	Gender					2.11	3.37	0.06	0.63		
	Puberty					-0.58	0.58	-0.12	-1.01		
	BMI					-0.72	0.45	-0.16	-1.60		
	Neuroticism					-1.96	0.34	-0.56	-5.81**		
Step 3		0.66	0.43	0.36	6.00(8,64)**					0.03	1.14(3,64)
	Constant					93.74	15.51		6.04**		
	Age					0.21	0.95	0.03	0.22		
	Gender					0.79	3.58	0.02	0.22		
	Puberty					-0.55	0.58	-0.11	-0.94		
	BMI					-0.69	0.46	-0.15	-1.52		
	Neuroticism					-1.97	0.34	-0.57	-5.87**		
	AuCg					23.15	13.38	0.47	1.73		
	AuCi					-27.89	20.56	-0.37	-1.36		
	Hair Cortisol					0.16	0.78	0.02	0.20		

Predicting Depression, Anxiety and Wellbeing Utilising Absolute Cortisol Variations

Table 4.4 demonstrates forced entry hierarchical multiple regression modelling, using the absolute cortisol concentrations from three time points (bed, wake and wake+30) to predict MFQ, SCAS and BBC. Control variables entered in the first block (age, gender, puberty and BMI) did not result in significant regression models in predictions of MFQ, SCAS or BBC.

The second step, involving the addition of neuroticism demonstrated a significant increase in r^2 for each of MFQ, SCAS and BBC, with significant beta coefficients in each case (see Table 4.4). The third step, entering of (log transformed) bed, wake and wale+30 cortisol values, demonstrated a significant increase in r^2 in models of MFQ (Δr^2=0.06, ΔF(3,65) =2.94p=0.04). However, this step did not demonstrate significant r^2 increase in predictions of SCAS or BBC. Beta coefficients of bed time cortisol concentration (β =0.20, t(72)=2.27, p= 0.03) were significant in relation to MFQ, in a positive direction. Standardised beta-coefficient's of waking concentrations were also significant in predictions of MFQ in a negative direction (β =-0.20, t(72)=2.14, p= 0.04); and positively in predictions of BBC (β =0.21, t(72)=2.00, p= 0.05).

Table 4.4: Hierarchical Regression Predicting Depression, Anxiety and Wellbeing using Absolute Cortisol Measurements (n=73)

Model	Predictor	r	r^2	Adj r^2	F-value (df)	Unstandardised Coefficients B	SE B	β	t-value	Δr^2	ΔF (df)
MFQ		0.27	0.07	0.02	1.32(4,69)					0.07	1.32(4,69)
Step 1	Constant					17.22	21.00		0.82		
	Age					-1.92	1.34	-0.20	-1.43		
	Gender					5.14	4.91	0.13	1.05		
	Puberty					0.64	0.83	0.11	0.77		
	BMI					0.76	0.65	0.14	1.18		
Step 2		0.69	0.48	0.44	12.64(5,68)**					0.41	53.86(1,68)**
	Constant					10.13	15.83		0.64		
	Age					-1.38	1.01	-0.14	-1.36		
	Gender					4.32	3.70	0.11	1.17		
	Puberty					0.06	0.63	0.01	0.10		
	BMI					0.31	0.49	0.06	0.63		
	Neuroticism					2.72	0.37	0.65	7.34**		
Step 3		0.74	0.54	0.49	9.68(8,65)**					0.06	2.94(3,65)*
	Constant					18.11	15.93		1.14		
	Age					-1.80	1.00	-0.19	-1.80		
	Gender					5.73	3.72	0.14	1.54		
	Puberty					0.02	0.61	0.00	0.03		
	BMI					0.45	0.49	0.08	0.92		
	Neuroticism					2.64	0.36	0.64	7.43**		
	Bed					5.50	2.43	0.20	2.27*		
	Wake					-16.89	7.90	-0.20	-2.14*		
	Wake+30					11.20	6.23	0.18	1.80		
SCAS		0.256	0.066	0.012	1.22(4,69)					0.07	1.22(4,69)
Step 1	Constant					9.34	29.19		0.32		
	Age					-1.91	1.87	-0.14	-1.03		
	Gender					4.43	6.83	0.08	0.65		
	Puberty					1.67	1.16	0.21	1.44		
	BMI					0.95	0.90	0.13	1.06		
Step 2		0.77	0.60	0.57	20.11(5,68)**					0.53	89.46(1,68)**
	Constant					-1.83	19.36		-0.09		
	Age					-1.06	1.24	-0.08	-0.85		
	Gender					3.14	4.52	0.06	0.69		
	Puberty					0.76	0.77	0.09	0.98		
	BMI					0.24	0.60	0.03	0.40		
	Neuroticism					4.28	0.45	0.74	9.46**		
Step 3		0.79	0.62	0.574	13.27(8,65)**					0.02	1.35(3,65)
	Constant					-12.58	20.14		-0.63		
	Age					-0.77	1.26	-0.06	-0.61		
	Gender					5.39	4.71	0.10	1.15		

	Puberty					0.80	0.77	0.10	1.04		
	BMI					-0.03	0.61	0.00	-0.04		
	Neuroticism					4.29	0.45	0.74	9.53**		
	Bed					-1.86	3.07	-0.05	-0.61		
	Wake					-8.69	9.99	-0.07	-0.87		
	Wake+30					-11.29	7.87	-0.13	-1.44		
BBC		0.31	0.10	0.04	1.81(4,68)					0.10	1.81(4,69)
Step 1	Constant					98.07	17.37		5.65**		
	Age					0.29	1.11	0.04	0.26		
	Gender					1.53	4.07	0.05	0.38		
	Puberty					-1.00	0.69	-0.21	-1.45		
	BMI					-1.04	0.54	-0.23	-1.94		
Step 2		0.63	0.40	0.35	9.00(5,67)**					0.30	34.24(1,68)**
	Constant					103.17	14.30		7.22**		
	Age					-0.11	0.92	-0.01	-0.12		
	Gender					2.12	3.34	0.06	0.64		
	Puberty					-0.58	0.57	-0.12	-1.02		
	BMI					-0.72	0.44	-0.16	-1.62		
	Neuroticism					-1.96	0.33	-0.56	-5.85**		
Step 3		0.66	0.43	0.36	6.41(8,64)**					0.04	1.67(3,65)
	Constant					98.97	14.77		6.70**		
	Age					0.28	0.93	0.04	0.31		
	Gender					0.73	3.45	0.02	0.21		
	Puberty					-0.50	0.57	-0.10	-0.89		
	BMI					-0.81	0.45	-0.18	-1.80		
	Neuroticism					-1.92	0.33	-0.55	-5.82**		
	Bed					-1.73	2.25	-0.08	-0.77		
	Wake					14.62	7.33	0.21	2.00**		
	Wake+30					-9.12	5.78	-0.17	-1.58		

Correlation Analysis of Cortisol, Emotion and Stressful Life Events

Due to power limitations relationships between emotional reactivity, regulation and stressful life events have been examined by Pearson's bivariate correlations employing bootstrapping and controlling for gender, age, puberty and BMI. Results are based on 1000 bootstrap samples. See Table 4.5 for full results.

The sensitivity subscale of the emotional reactivity scale demonstrated a significant negative correlation with waking cortisol concentration (r^2=0.12, p=0.01). Sensitivity, arousal intensity and persistence each demonstrated significant positive correlations with stressful life events (r^2=0.12, p=0.01; r^2=0.11, p=0.01; and, r^2=0.23, p=<0.001 respectively).

In terms of emotional regulation, reappraisal was significantly positively correlated with bed time cortisol (r^2=0.08, p=0.04), waking cortisol (r^2=0.21, p=0.001); AuCg (r^2=0.27, p<0.001); and, AuCi (r^2=0.11, p=0.02). Suppression was not significantly correlated with any cortisol measures or stressful life events.

Stressful life events were not significantly correlated with cortisol variables.

Table 4.5: Pearson's Correlations Depression, Anxiety, Wellbeing, Emotional Reactivity, Emotional Regulation, Cortisol and Stressful Life Events

		MFQ	SCAS	BBC	NEUROTICISM	SENSITIVITY	AROUSAL INTENSITY	PERSISTENCE	REAPPRAISAL	SUPPRESSION	BED	WAKE	WAKE+30	AuCg	AuCi	HAIR	STRESSFUL LIFE EVENTS
MFQ	r																
	Sig.																
SCAS	r	**0.73**															
	Sig.	<0.001															
BBC	r	**-0.74**	**-0.58**														
	Sig.	<0.001	<0.001														
NEUROTICISM	r	**0.70**	**0.77**	**-0.68**													
	Sig.	<0.001	<0.001	<0.001													
SENSITIVITY	r	**0.66**	**0.70**	**-0.63**	**0.85**												
	Sig.	<0.001	<0.001	<0.001	<0.001												
AROUSAL INTENSITY	r	**0.61**	**0.651**	**-0.47**	**0.80**	**0.90**											
	Sig.	<0.001	<0.001	<0.001	<0.001	<0.001											
PERSISTENCE	r	**0.74**	**0.77**	**-0.55**	**0.81**	**0.89**	**0.88**										
	Sig.	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001										
REAPPRAISAL	r	-0.11	-0.13	0.19	**-0.26**	**-0.34**	**-0.30**	-0.24									
	Sig.	0.44	0.35	0.17	0.06	0.01	0.03	0.08									
SUPPRESSION	r	**0.51**	**0.40**	**-0.40**	**0.35**	**0.32**	**0.32**	**0.40**	-0.08								
	Sig.	<0.001	0.003	0.003	0.01	0.02	0.02	0.003	0.57								
BED	r	0.23	-0.03	-0.11	-0.04	0.02	-0.01	0.02	**0.28**	0.19							
	Sig.	0.10	0.85	0.46	0.77	0.90	0.98	0.89	0.04	0.18							
WAKE	r	-0.18	-0.12	0.13	-0.11	**-0.35**	**-0.24**	**-0.27**	**0.46**	-0.11	-0.02						
	Sig.	0.20	0.40	0.37	0.42	0.01	0.08	0.05	0.001	0.43	0.88						
WAKE +30	r	0.02	-0.12	-0.17	-0.01	0.02	0.04	0.01	0.10	-0.03	**-0.36**	0.222					
	Sig.	0.87	0.41	0.23	0.93	0.90	0.76	0.94	0.48	0.85	0.01	0.11					
AuCg	r	0.11	-0.13	-0.08	-0.10	-0.17	-0.12	-0.13	**0.52**	0.09	**0.73**	**0.60**	0.15				
	Sig.	0.44	0.37	0.59	0.48	0.23	0.39	0.37	<0.001	0.52	<0.001	<0.001	0.30				
AuCi	r	-0.09	-0.15	-0.05	-0.07	-0.19	-0.11	-0.15	**0.33**	-0.08	-0.26	**0.73**	**0.83**	**0.45**			
	Sig.	0.54	0.30	0.75	0.61	0.18	0.45	0.29	0.02	0.56	0.06	<0.001	<0.001	0.001			
HAIR	r	-0.05	-0.07	-0.002	-0.002	-0.01	-0.08	-0.07	0.18	0.18	0.16	-0.10	0.10	0.12	0.02		
	Sig.	0.724	0.65	0.99	0.99	0.94	0.60	0.61	0.20	0.19	0.26	0.50	0.49	0.41	0.92		
STRESSFUL LIFE EVENTS	r	**0.52**	**0.38**	**-0.43**	**0.42**	**0.35**	**0.34**	**0.48**	-0.08	0.25	0.11	-0.06	-0.01	0.05	-0.05	0.03	
	Sig.	<0.001	0.01	0.002	0.002	0.01	0.01	<0.001	0.59	0.08	0.46	0.65	0.93	0.72	0.75	0.85	

Significant correlations are in bold; Correlation results are based on correlation analysis based on 1000 bootstrap samples; df=50

4.7 Discussion

This study examined the role of HPA axis activity on depression, anxiety and wellbeing as well as the role of emotional reactivity, emotional regulation and stressful life events on these variables. No relationships between our outcome measures of depression, anxiety and wellbeing and relative measures of daily cortisol fluctuations were demonstrated. Results indicate that absolute measures of cortisol at bed and waking were significantly predictive of depression (above that of neuroticism), but not of anxiety or wellbeing in adolescents.

This indicates that higher bedtime and lower waking cortisol were associated with increased levels of depression. Such pattern of higher bedtime and lower waking cortisol concentration may indicate blunted HPA-axis reactivity. Low evening cortisol levels are considered important for cueing sleep, while high waking levels are important for arousal and orientation necessary for wakefulness and day time functioning. This is in keeping with amotivational theories of depression and the theoretical explanations of the involvement of the HPA-axis (see Gold and Chrousos, 2002 for review). However, the relationships here were small and as such ought to be interpreted with caution. This may be due to the non-clinical nature of the sample and depression symptoms reaching non-clinical levels.

Previous research has identified that measures of cortisol concentration, particularly measures of cortisol awakening response (CAR), may represent biological markers for depression. This study calculated CAR based on AuC calculations and also included an overall AuC measure, reflecting total cortisol production (AuC_g). These measures were not found to be significant predictors of depression, anxiety or wellbeing scores. Although non-significant, reduced (or flattened) CAR was related to higher depression scores. This directionality supports previous research, which has associated a flattening of the CAR response with depression, particularly within adult samples. Research with adolescent samples has been somewhat inconsistent. For example, increased CAR, rather than reduced CAR, was prospectively predictive of depression in one study (Adam et al., 2010), while some research has demonstrated no association between CAR response and depression (Goodyer et al., 2000). Such inconsistencies may be a consequence of employing differing methodology and varying measures of cortisol. A strength of this study is that multiple measures of cortisol have been employed, yet no clear association has been demonstrated. Alternatively, it may be that cortisol measures are not robust predictors of depression within adolescents, perhaps due to developmental maturation. Keenan et al.'s (2013) indicated that there are age related changes in the sensitivity of the HPA-axis which may result in a delay of emergence of association

between measures of the HPA-axis and depressive symptomology. A further alternative is that dysregulation of cortisol regulation only occurs over time and recurrent exposure to depression (Keenan et al., 2013).

Although non-significant, the directionality of the relationship between cortisol and depression are in line with previous findings of reduced CAR in individuals with depression. It may be that due to the non-clinical nature of the sample that such a relationship has not been demonstrated. If, like theorised by Keenan et al. (2013), differences in HPA axis functioning is an accumulation of recurrent experience of depression, it may be that individuals within the current sample have not experienced this. Individuals within this sample may go on to develop depression and are in a premorbid state, or individuals who demonstrate clinically significant levels of depression may not have experienced the recurrent impacts of depression due to their age (mean age= 15.7).

It may also be that there is a more nuanced relationship between HPA axis activity and depression, anxiety and wellbeing than has been identified by this study. For example, assessment of cortisol may be helpful in differentiating between subtypes of depression, cortisol may reflect interaction between genes and environment. Cortisol production may also be particularly important when considering possible interactions with treatments for depression and anxiety. Herbert (2013) posited that depression that is resistant to serotonin-acting medications may be impeded by daily cortisol rhythms and that the addition of corticosterone may improve the effect of such treatment. If future research clearly identifies the role of cortisol within disorders, normalisation of HPA functioning may play an important role in treatments.

Two potential factors that may aid understanding of the HPA-axis' relationship with psychological health are emotional reactivity and emotional regulation. A second aim of this study was to examine any relationship between the emotional reactivity and regulation processes with the physiological stress-response system via cortisol concentrations. This study demonstrated significant relationships between measures of cortisol concentration and reactivity with measures of emotional reactivity and regulation. Higher emotional sensitivity was significantly associated with lower cortisol concentration measured at waking. It should also be noted that increased cortisol reactivity (both AuCg and AuCi) was significantly related to increase use of reappraisal as an emotional regulation strategy. As reappraisal is considered the more adaptive regulation strategy, it would make theoretical sense that it would be associated with greater reactivity of the HPA-axis as flattened or reduced responses have been

associated with poor physical and mental health outcomes. It may be that poorer emotional regulation results in detrimental alterations in the regulation of the HPA axis (or vice versa). This could have significant implications for both physiological and psychological health and is worthy of further study to more fully examine the relationships between these variables. Previous research has not addressed relationships between internal psychological mechanisms of emotional reactivity and regulation with the physiological mechanisms of stress response. This study supports the hypothesis that relationships between these components exist in adolescents. Importantly, the results of this study are in the direction that one would expect.

It is significant that psychological measures of emotional reactivity and regulation had much larger relationships with depression, anxiety and wellbeing than physiological measures. Although emotional reactivity and regulation do not measure psychological stress, they may be indicative of an individual's capacity to react to and regulate emotional stress (Shapero, Abramson and Alloy, 2015). Gunaydin, Selcuk and Ong (2016) demonstrated that reappraisal predicted lower measures of negative affect in response to daily negative events. As such, emotional regulation may be an integral component of coping with stress on a day-to-day level. This is tentatively supported by the findings of this study whereby measures of cortisol were associated with measures of reactivity and regulation. Consequently, these psychological measures may be more applicable in terms of identifying individuals at risk of developing psychological disorders compared to physiological measures of cortisol.

As expected and reiterating findings of the previous chapters, neuroticism demonstrated substantial predictive capacity in relation to depression, anxiety and wellbeing, accounting for 30-53% of symptoms. In addition, neuroticism was significantly associated with emotional reactivity, but not significantly related to emotional regulation strategy. This may indicate that neuroticism, a largely heritable trait, predisposes an individual to emotional reactivity. That it is not significantly related to regulation style may identify emotional regulation as a means that may be modified in order to improve coping. This is also of significance as regulation strategies were significantly associated with measures of cortisol. If one is able to improve emotional regulation it may be possible to improve psychological health and also mitigate any impact of psychological stress on physiological health.

Measures of stressful life events have previously been implicated in psychological disorders, this study supported these findings and demonstrated that stressful life events were significantly related to depression, anxiety, wellbeing, neuroticism, emotional sensitivity, emotional arousal intensity and emotional persistence (~12-27% shared variance). This is of

clinical relevance as stressful life events are considered to increase risk of disorder onset and severity. It is also significant is that emotional reactivity variables were associated with stressful life events. This may indicate that emotional reactivity is impacted by experiencing stressful life events. This has the potential to impact the emotional experience of future events which may further increase the risk of disorder. However, due to the cross-sectional nature of this study, definitive conclusion regarding causal relationships cannot be drawn. Longitudinal studies may address this and tease apart these factors to examine their impact on depression, anxiety and wellbeing. Nevertheless, measures of emotional reactivity, emotional regulation and stressful life events may be a useful way to identify individuals at risk of psychological disorder, facilitating early intervention.

These findings highlight the importance of emotional reactivity and regulation in symptoms of depression, anxiety and wellbeing. Additionally, relationships between emotional reactivity and regulation were demonstrated with HPA-axis activity. This may have significant clinical implications in that the experience of emotions may be modifiable by treatment which may improve psychological outcomes. As the HPA-axis has been implicated in poor outcomes for physical health it is important to consider potential means of reducing this effect. If emotional reactivity and regulation play a role in the activity of the HPA-axis, they may be key targets for intervention. Further research to identify the direction of the relationships is needed to more fully understand the relationship between these components prior to the development of any modification intervention.

A strength of this study has been the broad assessment of psychological health. Diagnostic categories have not been employed in order to reflect the concept of mental health and wellbeing being comprised of a spectrum rather than distinct categories. This approach is less frequently employed within research. Additionally, recruiting this sample from mainstream schools was designed to ensure that the sample is more representative of the general population of adolescents. As previously discussed, recruiting only from clinical settings risks failing to capture individuals that are not help seeking or those who fall below diagnostic cut-off points. Although it is important for research to address these clinical groups, overreliance upon such samples in turn presents the problem of skewing research findings to a narrower group, which may result in a gap of understanding.

There are some limitations within this study. Firstly, all psychological measures were assessed through self-report questionnaires, which might be subject to responder biases. An additional

challenge throughout the data-collection phase was ensuring saliva samples were deposited at the requested time as well as the returning of completed saliva sampling kits. While every effort was made to explain why the timing of giving the sample and the recording of that time was of importance, the degree to which these instructions were followed by participants has not been assessed. As such, it is possible that variation exists in the extent to which individuals complied with these instructions. There may also be an element of social desirability bias, whereby participants who may have forgotten to complete samples at the appropriate time did so outwith the requested time. Some individuals failed to return saliva samples, which has contributed to missing data. All efforts were made to arrange to collect samples at an alternative time. Efforts to minimise the impact of these problems on the findings were also made. For example, samples that did not contain a time of collection were excluded from analysis as were those if the recorded wake + 30 deposit time was more than 40 minutes after the initial (wake) sample. These exclusions have inevitably contributed to missing data. It is positive to note, however, that the examination of overall cortisol production patterns demonstrated the expected pattern, illustrating the reliability of the data that has been included. While repeated measures remain an optimal design within studies of this nature, in practice collecting many samples and ensuring the validity and reliability of these samples presents logistic difficulties.

Secondly, the assessment of CAR has relied upon a two-sample protocol. While this has been employed within previous research (e.g. Adam et al., 2010), more intensive protocols (e.g. sampling every 10-15 minutes) are likely to be more precise and identify more subtle changes in CAR. Additionally, single day sampling is limited and means of multiple day samples would be preferable in terms of reliability (Helhammer, Fries, Schweisthal, Schlotzz, Stone and Hagemann, 2007).

An additional issue relating to analysis of cortisol concentrations within samples, both saliva and hair samples, is that biological or technical replicates of analyses were not conducted due to the weight or volume of the sample produced, as well as the financial cost. Nevertheless, the variability within and between samples was low. An additional implication of the low variability is that differences between participants would not necessarily be expected. Future work may consider using repeated sampling over a longer period of time and using mean scores of cortisol concentrations for further analysis.

Additionally, the relationships assessed here have assumed a linear relationship between variables. It may be that the impact of cortisol, or indeed other examined relationships, is not linear. If this is the case, then logistic regression analysis may be more appropriate. Furthermore, the effect of some variables (puberty and medication use) have not been fully assessed due to categorical nature and sample size limitations.

Finally, sample size has limited the present study. The final sample size (due to missing data) fell below the desired number. This has resulted in analyses being underpowered and as such, may have failed to detect significant relationships. Larger sample sizes could be employed to more fully examine these relationships. As the relationships demonstrated within this study are small, investigation of these relationships within a substantially larger sample is warranted. Although attempts were made to recruit equal numbers of males and females, as is the case within much psychology research, males are underrepresented within this sample. Furthermore, the fact that multiple tests have been conducted within this analysis has increased the likelihood of a type II error. Efforts to minimise this, such as employing p-level adjustments for multiple comparisons, have been made; nevertheless, results ought to be interpreted with caution until they are replicated in the future.

4.8 Conclusion

Overall this chapter implicates stressful-life-events and the emotional processes of reactivity and regulation in symptoms of depression, anxiety and wellbeing in adolescents. As such, individuals experiencing stressful-life events may be considered at increased risk of disorder, particularly within the context of the personality trait neuroticism and emotional processing abilities. Future work may examine interaction between these variables. Additionally, further exploration to increase understanding of the role of the HPA axis is warranted. Results demonstrated that these relationships are small and that cortisol measures did not significantly increase the ability to predict depression, anxiety and wellbeing. However, some small but significant relationships were exhibited, partially supporting the relationship between psychological health and HPA-axis functioning as well as, implicating emotional reactivity and emotional regulation in HPA-axis activity and psychological health. It is possible that a larger sample size would have increased power to identify such effects. These findings support the strong role of environmental influences in addition to personality influences, as well as the smaller impact of HPA axis regulation. Importantly, psychological measures of emotional regulation and reactivity demonstrated stronger relationships with depression, anxiety and

wellbeing than physiological measures of the HPA axis. The association between emotional reactivity and regulation may implicate these emotional processing factors in the activation of the physiological stress response system, this would have significant clinical implications. Future studies may consider examining these factors in a prospective longitudinal design with a large sample size in order to more fully unpick the nature and directionality of these relationships.

Chapter Five – Do subjective and objective measurements of sleep predict depression, anxiety and wellbeing?

5.1 Introduction

As discussed in previous chapters, adolescence represents a period of vulnerability for the onset of mental health concerns as well as a potential window of opportunity to nurture positive wellbeing that may have lifelong impact. This period of sensitivity is partly attributed to brain development and associated plasticity. Research suggests that normal brain and body homeostasis is dependent upon sleep (Siegel, 2005; Tarokh, Saletin and Carskadon, 2017). Neural development and reorganisation occurring during adolescents is associated with substantial change in sleep architecture. Brain development is particularly sensitive to sleep; although long-term effects are unclear within humans, animal studies demonstrate that sleep is critical for elements of neural development (Dahl, 2007). Furthermore, within human studies sleep has been strongly linked to cognitive performance. As such, sleep is likely to be of increased importance during periods of increased development, like adolescence.

However, adolescents' sleep tends to fall below the National Sleep Foundation's 8-10 hours per night recommendation (Hirshkowitz, et al., 2015). Evidence, such as an increase in daytime sleepiness and increased sleep duration on weekends (Tarokh, Saletin and Carskadon, 2017) has been related to socially imposed early waking times (i.e. school start times). This evidence suggests that the reduced duration of sleep during adolescence is socially rather than biologically constrained. Sleep has also been shown to be of importance in maintaining wellbeing and psychological health (Telzer, Goldenberg, Fuligni, Lieberman and Galvan, 2015). This is of particular concern as it coincides with the sharp increase in instance of psychiatric disorders during this developmental stage (Kessler, Berglund, Delmer, Jin, Merikangas and Walters, 2005). It is also significant that problems with sleep feature heavily in diagnostic criteria for many psychiatric disorders. The focus of this chapter is to consider sleep behaviour in the context of adolescent development and psychological health. Initially discussing the literature background of this area, focusing on evidence linking sleep with depression, anxiety and wellbeing, as well as, vulnerability by virtue of neuroticism, in the adolescent population. It will then consider the potential role of emotional reactivity and

emotional regulation. Finally, an empirical study examining these factors will be described and discussed.

The Sleep, Arousal and Circadian Timing

Evolution has resulted in adaption to the daily variation in temperature and light. The circadian (from the Latin *circa,* meaning 'about' and *dies,* meaning 'day) clock is an internal mechanism, maintained by the suprachiasmatic nucleus (Reinberg and Ashkenazi, 2003), that is synchronised to the ~24-hour light/dark cycle (Panda, Hogenesch and Kay, 2002). The circadian clock regulates various elements of physiology and behaviour, research and understanding of its actions and associated mechanisms are a major focus of current research, knowledge of its effects is expanding, although understanding is far from complete. The circadian clock has demonstrated a regulating influence on biological activity (e.g. gene expression and tissue generation), as well as, controlling behaviour (e.g. sleep and arousal) in both human and non-human species (Panda et al., 2002).

Human sleep patterns are partly regulated by the circadian clock resulting in a pattern of wakefulness during daylight hours and sleep during darkness. However, variation in preference sleep timing exists in humans, referred to as 'chronotype', which has been shown to be regulated largely by the circadian clock and coded by genes (Kalmbach, Schneider, Cheung, Bertrand, Kariharan, Pack and Gehrman, 2017). Chronotype signifies an individuals' preference to 'morningness' or 'eveningness' (Barclay, Eley, Parsons, Willis and Gregory, 2013). Hormones, regulated by circadian timing, are mechanisms that induce sleep and arousal. Typical hormone profiles show that melatonin (signalling sleep) builds gradually throughout the day and peaks in the evening predicting sleep onset (Zamanian, Dehghaani, and Hashemi, 2013). At the same time, cortisol (cueing arousal) increases sharply in the morning, peaks approximately 30 minutes following waking (known as cortisol awakening response) then falls gradually throughout the day (Zamanian et al., 2013).

A second process that interacts with the circadian regulation to regulate sleep is the homeostatic process of the accumulation of sleep pressure. Sleep pressure (sleepiness) builds during wake and is impacted by the time spent awake and the duration of prior sleep (see Barclay and Gregory, 2014, for review). Jenni and LeBourgeois (2010) provide a review of sleep behaviour throughout childhood and adolescence in relation to this bioregulatory model.

Adolescent Circadian Preference

Crowley, Acebo and Carskadon (2007) demonstrated that circadian phase preference of evening chronotype was correlated to pubertal development in adolescents; that older adolescents, who had greater pubertal development, demonstrated later circadian phase preferences than younger, less developed, adolescents. Furthermore, Jenni et al and LeBourgeois (2010) demonstrated that sleep pressure accumulated more slowly in older adolescents compared to pre-pubertal adolescents, as such, pre-pubertal adolescents demonstrated quicker sleep onset. This suggests that more developmentally mature adolescents are more able to stay awake for longer periods of time and delay sleep onset and that this is associated with the shift to evening chronotype. A move in chronotype away from the evening type and towards the morning type has been referred to as a marker of the end of adolescence. Roenneberg et al., (2004) considers typical chronotypes to reach peak lateness at around the age of 20, following which individuals demonstrate a gradual return to earlier chronotype, while retaining individual differences (Roenneberg, et al., 2004). As such, chronotype may be a useful tool in categorising adolescence.

A biologically driven circadian shift to later chronotype is associated with the adolescent period. This has been considered to result in 'social jet lag' due to social constraints of conventional wake times (Wittmann, Dinich, Merrow and Roenneberg, 2006). This is thought to contribute to the sleep deprivation experienced by adolescents. Shifting between early rising times during the week and the ability to sleep longer during the weekend, may leave adolescents unable to fully synchronise their sleep-wake behaviour to fit with social constraints (see Carskadon, 2002 for review). The abrupt shifting between the two limits the internal circadian clocks ability to adjust, resulting in a vicious cycle of sleep deprivation and tension between sleep needs and circadian constraints.

Environmental factors have also been considered to influence chronotype. In order to examine environmental factors while excluding genetic influences, Barclay et al. (2013) conducted a study of 189 late adolescent monozygotic twin pairs (mean age 19.81). When controlling for genetic and shared environmental influences, differences in chronotype were associated with negative life events, educational attainment, smoking and drug use. Chronotype may enact a causal influence over behaviour. If late chronotype results in insufficient sleep this may negatively interfere with daytime functioning which may in turn have long-term deleterious consequences. Consequently, individuals may resort to adopting maladaptive coping

strategies, such as smoking and drinking which could exacerbate negative effects (Urban, Magyarodi and Rigo, 2011). Similarly, drug use and smoking may be considered symptomatic of poor coping with social demands; or, are simply behaviours that are associated with increased evening activity that late chronotypes engage in (Steinhausen and Metzke, 1998). These findings suggest that adolescents and young adults would greatly benefit (possibly with positive life-long consequences) from a greater understanding of these biological systems.

Sleep and Mental Health in Adolescence

The National Sleep Foundation considers adolescents to be chronically sleep deprived and found that almost half of adolescents had insufficient sleep (2000). This is important as sleep reduction in adolescents has been associated with a variety of detrimental outcomes including poor school performance, high risk for emotional disorders, obesity, cardiovascular problems, risk-taking behaviour, car accidents and physical injury (e.g. Danner and Phillips, 2008). Furthermore, prospective data collected from a community-based sample, demonstrated that levels of short sleep (<6 hours) comprised an increased risk for symptoms of depression (Roberts and Duong, 2015).

Sleep disruption is a key diagnostic criterion of depression and anxiety (APA, 2013). Roberts, Lewinsohn and Seeley (1995; in Ivaneko, Crabtree and Gozal, 2005) conducted a large-scale longitudinal study of sleep and emotional disorders in adolescents. They found that while 2.6% of individuals met clinical criteria for depression, 88.6% of those who did experienced sleep disturbance. Even more strikingly, 75% of those who went on to develop depression over the follow-up period (13.8 months) had previously reported sleep problems (Ivaneko et al., 2005). It may be that disrupted sleep constitutes a preclinical or early symptom of disorder, or that it plays a role in the onset and development of disorder. Furthermore, a recent meta-analysis, found that adolescents with depression had longer sleep onset latency (d=0.27); more awakenings (d=0.44); and, lower sleep efficiency(d=-0.38), compared to those who were not depressed or those who had undergone remission (Lovato and Grandisar, 2014). These findings indicate that wakefulness while in bed is a key factor differentiating those that are depressed from those who are healthy or in remission.

Sleep loss has been demonstrated to increase negative affect in animal models as well as mediating the relationship between physiological stress and depression in humans (Novati, Roman, Hagewoud, Boer, Luiten and Meerlo, 2008; Hamilton, Catley and Karlson, 2007).

Recent genetic research also substantiates the association between depression and sleep symptoms. Wray et al., (2017) found positive genetic correlations with daytime sleepiness, insomnia and tiredness. This finding supports a strong mechanistic connection between sleep and depression. Sleep problems may constitute an important factor, Riberio, Pease, Guiterrez, Silva, Bernet, Rudd and Joiner (2012) demonstrated that sleep problems were the best predictor of suicidal ideation, outperforming other risk factors including depression, hopelessness, PSTD, anxiety, drug and alcohol abuse ($pr = .12, t\,[307] = 2.11, p < .05$).

Another line of inquiry has been to investigate the impact of treatment for psychiatric illness on sleep. Lemoine, Gilleminault and Alvarez (2007) demonstrated that antidepressant treatment was related to significant improvement in measures of sleep. Of note is that significant improvements in subjective sleep measurements were observable from the first week of treatment; whereas, improvements in depressed mood have a longer onset period.

Despite findings of association and the predicative value of sleep measures in the instance of psychiatric disorder, there is a lack of consensus on the specificity of any particular sleep feature in distinguishing between disorders (Benca, Obermeyer, Thisted and Gillin, 1992). As such, Harvey, Murray, Chandler and Soehner (2011) propose sleep disturbance to be a transdiagnostic mechanism associated with psychiatric disorder. Harvey et al., (2011) posit that sleep disturbance is aetiologically related to psychiatric illness through emotional regulation and the dopaminergic and serotonergic systems.

Sleep and Personality Risk

Investigations of sleep and risk for depression has resulted in examination of neuroticism in the context of sleep behaviour. Chronotype and neuroticism have been identified as having considerable genetic underpinnings and have been examined in relation to each other. For example, Kalmbach et al., (2017) conducted a large-scale genome-wide association study, to identify chronotype as a heritable trait associated with nine gene variants. Randler (2017) demonstrated that it correlates with developmental markers and personality type (Randler et al., 2017). Neuroticism, a largely heritable personality trait and risk factor for depression (e.g. Power and Pluess, 2015), was related to evening chronotype, as well as, longer sleep latency and longer sleep duration on weekends in a large nationwide adult sample. Furthermore, studies of young adults demonstrate high neuroticism to be predictive of subjective sleep quality (Cellini, Duggan and Sarlo, 2017). Additionally, in individuals with insomnia,

neuroticism was found to be the best predictor of subjective sleep quality. Subsequently, chronotype preference, sleep latency, duration and quality may constitute markers of predisposition or risk of disorder.

While neuroticism has been the subject of some research in relation to disordered sleep and psychological disorder, the majority of research focuses on clinical disorders and sleep excluding personality risk factors (Le Blanc et al., 2007). Few studies have examined neuroticism and both objective and subjective measures of sleep particularly within adolescent samples. Gau (2000) found that high neuroticism was related to subjective measures of later bed times on school nights, reduced total sleep time, greater feelings of sleep insufficiency, tiredness, moodiness and difficulty rising. As part of a longitudinal study, Danielsson, Jansson-Frojmark, Linton, Jutengren and Stattin (2010) found that neuroticism measured at age 16 was not predictive of subjective sleep-onset problems later, but rather, that subjective sleep-onset problems were predictive of neuroticism at age 37.

A systematic review concluded that the best evidence indicates a bidirectional relationship between disrupted sleep and emotional disorders; but that studies of childhood and adolescent disorders found sleep problems to be predictive of onset of anxiety and depression (Alvaro, Roberts and Harris, 2013). Although variation of findings exists, this evidence indicates that poor sleep may be a marker of emotional disorders and constitute an important, and identifiable, vulnerability factor for the onset of disorders. Associations between sleep disturbance and mood disorders are well established in adulthood; however, fewer studies examine this relationship in adolescence. A recent review indicated inconsistencies of findings, particularly relating to subjective versus objective measurement of sleep behaviour (Gregory and Sadeh, 2012). As adolescence is a unique developmental phase associated with associated with major changes in sleeping patterns (Urrila, Puanio, Palomaki and Marttunen, 2015) as well as a sharp increase in the prevalence of mood disorders (Thapar, Collishaw, Pine and Thapar, 2012), it is of key importance that adolescents are studied separately from children and adults.

This is an important line of research as sleep preferences and behaviour are relatively easily identified and therefore may provide a straightforward and unobtrusive means of assessing risk factors. Furthermore, stigma surrounding mental illness and wellbeing, is known to be a barrier to discussion and accessing support (Gulliver, Griffiths and Christensen, 2010). If sleep problems are less stigmatised than psychiatric symptoms, this may provide an alternative entry

point to facilitate identification of psychological difficulties and risk thereby improving access to support.

Sleep and Emotion

Emotional processing has been identified as a potential mechanism driving the relationship between sleep and depression (Harvey et al., 2011). Within the current context, emotional regulation has two important components: emotional reactivity, which is considered the experience and generation of emotion; and, emotional regulation, the strategies, employed by individuals to cope with emotions experienced. Recent review by Palmer and Alfano (2017) concludes that disrupted sleep is a robust risk and maintenance factor for various psychiatric conditions and identify emotional regulation as a potential mechanism by which sleep both impacts upon and is impacted by psychological outcomes. Substantiating this, Gregory and Eley (2005) demonstrated a correlation (r=-.38) between negative attributional style and sleep problems in children aged 8-11. Additionally, Alfano, Zakem, Costa, Taylor and Weems (2009) demonstrated correlations between sleep problems and depression (r=0.58) and anxiety (r=.41) in a sample of adolescents recruited from the community.

Evidence from neuroscientific studies indicates that emotional regulation involves the interaction of bottom-up processes of the limbic system (e.g. amygdala) which is implicated in the generation of emotional responses, emotional reactivity and behaviour with the top-down regulatory processes of the prefrontal cortex that are responsible for regulating emotional responses and behaviour (Palmer and Alfano, 2017). Individual differences are exhibited in both reactivity and regulation of emotions. Evidence indicates that the structures and neurochemicals involved in emotional reactivity and regulation are also implicated in the governance of sleep (Goldstein and Walker, 2014).

However, research investigating the role and relationship between emotional regulation and sleep are extremely limited in number (Palmer and Alfano, 2017), despite much interest within the research area of the bidirectional relationship between sleep and emotion. Overall, evidence discussed in recent review by Palmer and Alfano (2017) suggests that emotional arousal and distress can cause difficulty falling asleep and sleep disruption and that sleep disruption can have negative emotional effects. Given these bi-directional effects, and that sleep, as well as, emotional regulation and emotional processing are implicated in psychological disorders, it may be expected that the brain areas recruited by emotional

processing and regulation are also implicated in sleep regulation. It could be predicted that psychological disorders and sleep disruption frequently co-occur, have bidirectional impact and may be mutually reinforcing.

Yoo, Gujar, Hu, Jolesz and Walker (2007) conducted an experimental manipulation to examine neurological changes as a consequence of sleep. Twenty-six individuals (mean age=24.1) were assigned to either sleep as normal or sleep deprivation conditions prior to an emotional viewing task. As expected, amygdala activation was demonstrated in response to increasingly negative stimuli. However, the sleep-deprived group demonstrated 60% increase of amygdala activation compared to the control group. Sleep-deprived individuals demonstrated greater amygdala connectivity with autonomic-activating brainstem centers (locus coeruleus and midbrain). Whereas, the control group demonstrated significantly stronger connectivity between the amygdala and the medial prefrontal cortex. These findings indicate that sleep deprivation initiates a hyper-limbic response by the amygdala in relation to negative emotions, which was also related to a loss of functional connectivity with the medial prefrontal cortex. This suggests that there is a failure of top-down prefrontal cognitive control following sleep deprivation. This finding is of significance as the prefrontal cortex has been widely implicated in emotional regulation (e.g. Ochsner and Gross, 2008) and emotional disorders (e.g. Drevets, 2001).

Studies examining these factors lack consistency of findings. For example, contradicting the previously mentioned studies, using subjective measurements within a young adult majority female sample (mean age 26.6), Cellini, Duggan and Sarlo (2017) found that emotional regulation strategies were not predictive of sleep quality (r^2=.01) in multiple regression analyses. Research in this area is also limited by the large reliance upon subjective measurements of sleep, which may not be an accurate estimate of sleep and is likely to be subject to response bias and influenced by factors such as daytime sleepiness and fatigue which may not be attributed solely to quality or quantity of sleep. Furthermore, sleep quantity tends to be the primary focus, leaving measures of quality and disruption less examined.

As highly arousing states are likely to conflict with the sleep, one might expect individuals experiencing high intensities of emotion to experience disrupted sleep. Talbot, McGlinchey, Kaplan, Dahl and Harvey (2010) examined the relationship between mood, sleep and emotional regulation strategy under conditions of sleep deprivation or optimal sleep. Results indicated that the deprivation condition was associated with reduced positive affect, increased

anxiety during a catastrophising task and interpreting threats as more threatening. Although emotional regulation strategies were not explicitly measured in this study, the finding that worry and anxiety increase due to sleep deprivation may indicate that sufficient sleep is necessary for the ability to successfully regulate emotion. The adolescent period has been associated with the subjective experience of high intensity of emotion. This is considered to be related to the relative under development of the prefrontal cortex in relation to limbic brain structures. As the prefrontal cortex is implicated in employing emotional regulation strategies which are considered to mitigate or control the impact of emotions, adolescents may be disproportionately affected by the impact of emotions.

Initial evidence of overlap and interplay between emotional arousal and regulation strategy warrants further study given its potential importance in the role of psychological disorders as well as the significance of sleep difficulties in psychological disorders. Disordered sleep is a prominent factor and its identification may improve the detection of psychological disorders and comprise a target for early intervention. If sleep may be improved, there may be multidimensional benefit to both physical and psychological health.

Objective vs Subjective Measurements

Sleep quality was found to be more related to health, life satisfaction, depression and fatigue than sleep quantity (Pilcher, Ginter and Sadowsky, 1997), in a sample of college students (mean age 20.9 years). Subjective reports of sleep suggest that depressed individuals feel that they have a reduced quality of sleep. Subjective measures of insomnia, hypersomnia, sleep quality and sleep disturbance all demonstrated significant difference between the two groups (Lovato and Gradisar, 2014). This meta-analysis indicates that subjective measures may be more able to distinguish between depressed and healthy groups. The discrepancy between objective and subjective measures may be that studies or methods are not sensitive enough to detect differences or that differences may be related to specific subtypes or phenotypes of disorder, which has not been fully explored by previous research (Gregory and Sadeh, 2016).

Alternatively, subjective reports of increased problems may be a consequence of mood bias within the groups; subjective measures may reflect depressed mood rather than poor sleep. It may be that depressed individuals' reports of sleep demonstrate a mood-congruent bias. Bertocci, Dahl, Williamson, Iosif, Birmaher and Axelson et al., (2005) demonstrated that 8-17-year olds with depression did not demonstrate any evidence of disturbance compared to

healthy control using polysomnography measurements, despite significant differences in self-report measures of sleep quality. A meta-analysis found sleep disturbance in adolescents to be predictive of the onset of depression (Lovato and Gradisar, 2014). Studies included in this meta-analysis comprised a mixture of objective and self-report measures of sleep problems. Objective measures included: sleep onset latency, wake after sleep onset, number of awakenings, time in bed, and sleep efficiency. Overall conclusions of relationships between these measures are limited due to the limited number of studies employing these measures. Of these measures, only the number of awakenings and sleep efficiency demonstrated significant difference ($p<0.05$) between depressed and control groups.

Furthermore, examination of polysomnography studies relating to stages of sleep and wave density indicated that when comparing depressed and control groups, significant differences were present only in density of Rapid Eye Movement sleep. However, Dahl and Lewin (2002) report that the majority of studies report negative findings here. Similarly, review by Gregory and Sadeh (2012) of studies using polysomnography, failed to find differences in children and adolescents with depression compared to controls. Gregory and Sadeh (2012) conclude that the evidence associating objective differences of sleep within depressed groups is unsupported by objective measures. Further investigation into the validity of subjective measures and the correlation between subjective and objective measures of sleep may help disentangle this relationship.

Measurement Techniques

Self-Report Measure: Diaries and Questionnaires

Self-report measures, including retrospective questionnaires and sleep diaries are frequently employed within research. Sleep diaries are considered the 'gold standard' of subjective measures of sleep (Gregory and Sadeh, 2012). However, both sleep diaries and questionnaires are associated with some significant limitations. For example, questionnaires may ask individuals to recall features of their recent sleep, which may be subject to recall bias, errors and response bias. Furthermore, when examining mood in relation to sleep, correlations between the two measures may be due to a reporting bias as a consequence of either/both depressed mood and sleep difficulties.

Objective Measures: Polysomnography

Polysomnography, recording heart rate, eye movement and which includes electroencephalogram, is the gold standard for the measurement and assessment of sleep.

However, polysomnography is invasive, expensive and typically relies on assessment of individuals in a sleep laboratory. As such, various other measures have been devised. Self-report measures assessing the quality and quantity of sleep are widely used and include self-report scales as well as sleep diaries. These measures assess various elements of sleep including time taken to fall asleep, length of sleep, satisfaction with sleep, night-time waking and daytime sleepiness. One problem with self-report scales is that some lack poor discriminant validity, due to items failing to capture incremental increases of symptom severity. Furthermore, to some extent, individuals may not be aware of various facets of their sleep such as restlessness and awakenings.

Objective Measurements: Actigraphs

Actigraphic devices allow for the unobtrusive measurement of sleep based on measures of accelerometetry and (depending on the device) other measures such as light exposure or heart rate. These devices have been used in research and have been employed as diagnostic tools for medical conditions (e.g. sleep apnoea, Garcia-Diaz et al., 2007). Actigraphs are typically small activity monitors that can be worn as wristwatches. Lichtenstein's (2006) validation study found that the Mini Mitter Actigraphy device slightly overestimated the number of awakenings, total sleep time and sleep efficiency while wake after sleep onset was slightly underestimated relative to polysomnography. However, the data recorded with by actigraphy were much more similar to that of the polysomnography than self-report sleep diaries. Overall, Lichstein (2006) concludes that actigraphy is acceptable for the clinical evaluation of insomnia in all elements of sleep other than sleep onset latency where correlations were weak; although the authors explain that this is likely due to the clustering of data across participants between 0 and 30 minutes. The inconsistent measurement of this element of sleep demonstrated in this study leant towards the actigraphy device slightly underestimating sleep onset latency, thereby recording a better sleep than the polysomnography. This validation study however, was conducted nearly ten years ago, and it is therefore likely that the technology has improved and so the recording of sleep will be even more effective.

One significant problem is that Lichstein (2006) found that the same robustness of findings was not found in relation to insomnia patients' relative to previous studies; previous studies have demonstrated correlations of r=.8-.9, as such this may be an artifice of this particular study (the specific device used) or this population more generally. Tyron (2004) similarly proposed that actigraphic measurements of sleep to be problematic in the case of insomnia due

to overestimation of sleep time due to the devices recording awake, but inactive participants, as asleep due to their failure to exceed the movement threshold for wakefulness. Further, Tyron (2004) highlights that polysomnographic measurements of sleep are associated with sleep onset stage phase two (reduced muscle tension) whereas, actigraphic measurements are associated with sleep onset stage phase one (quiescence), thereby accounting for the discrepancy between sleep onset latency and sleep duration between actigraphy and polysomnography.

Since Lichtenstein's (2006) study and Tyron's (2004) review, various researchers have utilised actigraphic devices to assess sleep in numerous populations, with predictive success. Avery, LeBourgeois, Gupta and Mittal (2015) recorded with an Actigraphic device, 38 adolescents (age 12-21 years) considered ultra-high-risk for the development of psychosis, as well as 31 controls, and demonstrated the ultra-high-risk group to experience an increase in wake time after onset, increased movement during sleep and decreased efficiency of sleep compared to healthy controls. Furthermore, trend-level associations between Actigraph and self-report (Pittsburgh Sleep Quality Index, Buysse et al. 1989) indices of sleep duration and efficiency were found. However, there was no association between awakenings by these two measurements indicating either that individuals are less able to self-report number of awakenings due to impaired recall of less conscious states or that Actigraphic devices were greatly over estimating awakenings. Importantly, the Actigraphic data of sleep disturbance predicted the longitudinal course of symptoms over a 12-month follow-up period in the ultra-high-risk group. This perhaps indicates that the Actigraph is more reliable and valid than self-report measures of sleep as it is able to record information that is not captured by self-report measures.

Support for the use of Actigraphy is also derived from an epidemiological study examining sleep and wellbeing (Lemola, Ledermann and Friedman, 2013). Lemola et al. (2013) demonstrated that in both white and African Americans, high day-to-day variability, as measured over a seven-day period, was associated with lower levels of self-reported wellbeing even after controlling for age, gender, education level, marital status and BMI. Whereas, sleep duration, sleep onset latency, and time awake after sleep onset were found to not be related to subjective wellbeing after controlling for covariates and other sleep variables. Furthermore, the relationship between variability in sleep duration and wellbeing was partially mediated by subjective experience of sleep quality. As such, this study indicates that variability in sleep duration is more related than average sleep duration is related to poor subjective sleep quality

and wellbeing. Lemola et al., (2013) utilised the Mini Mitter Actiwatch and Actiware 5 Software with manufacturer algorithms for detecting sleep based on 30 second epochs to infer details of sleep elements, a formula previously validated by Oakley (1997).

While polysomnography remains the gold standard of sleep measurement, in the absence of polysomnography, actigraph measurements may provide valid objective measurements, P particularly when considering that technological advancements have improved the ability of such devices to accurately estimate various elements of sleep. Actigraphy comprises a useful tool in the measurement of sleep, particularly elements that are less validly assessed through self-report questionnaires, such as the efficacy of sleep and number of awakenings experienced.

Discrepancies have been found between self-report measures and Actigraph data; however, it is possible that this is not solely due to unreliability of the Actigraph data but a lack of validity of the self-report scales. Ancoli-Israel, Cole, Alessi, Chambers, Moorcroft and Charles (2003) report that it is commonplace for individuals to complete sleep diaries for multiple days at one time rather than every day, furthermore, it is also possible that these measures are subject to reporting bias. As such, actigraphy can provide an objective measure of sleep to use in conjunction with self-report measures to give a more reliable overall picture of sleep. A comprehensive review established that Actigraph devices were useful in the measurement of sleep, particularly when used in conjunction with self-report measures (Ancoli-Israel et al., 2003). Ancoli-Israel's (2003) review concludes that actigraphy is more reliable than sleep logs, can record data continuously for long time periods and are able to capture sleeping behaviours in situ thereby demonstrating high ecological validity.

Meltzer, Walsh, Traylor and Westin (2012) evaluated the reliability of the Actiwatch 2 in a child and adolescent sample, to indicate that this approach demonstrate no significant differences in total sleep time compared to polysomnography recording, an improvement on other comparable devices. However, the software overestimated wake after sleep onset by ten minutes and a 0.4% underestimation of sleep efficiency. Meltzer et al. (2012) conclude that these differences although statistically significant are sufficiently small that they do not reflect clinically meaningful differences.

5.2 Rationale

Inconsistencies within research findings, methodological variations within previous research and limited research within adolescent populations mean that further study investigating the relationships between sleep, psychological health and potential mechanisms of any relationships is warranted. High levels of low mood within a sample recruited from community settings were demonstrated in previous chapters. As such, it is important for research to target this community group as evidently many young people experience low mood but are not necessarily help seeking and so are unlikely to be well represented in a clinical sample. Dahl and Lewin (2002) propose that this area is under researched and further study is required to describe links between sleep, behaviour, psychopathology and affect regulation Consequently, this study aims to examine the relationship between sleep and emotional regulation, and relationships with symptoms of depression, anxiety and wellbeing, as well as the risk factor neuroticism.

5.3 Hypotheses

Based on previous research, it is predicted that objective and subjective measures will be related to distinct components of psychological measures and that subjective sleep measures will be superior predictors of depression, anxiety and wellbeing, compared to objective measures. Furthermore, it is predicted that emotional reactivity and regulation will be related to both sleep measures and psychological measures. It is expected that measures of reduced quality and quantity of sleep will predict an increase of depression and anxiety symptoms as well as a decrease in wellbeing.

5.4 Methodology

Data for this study were collected in conjunction with the previous study (Chapter Four). Details regarding participant recruitment and procedure of this study have been described in full in Chapter Four. The following section will briefly summarise the methodological details and describe the measures of sleep. Combining the data collection element of these studies was considered optimal for the examination of the rich source of data within a single sample.

Participants

Data were collected from 132 participants over the course of one week, 74 of whom wore Aactiraph devices (one participants data was unrecoverable due to a technical problem), 93

participants completed and returned sleep diaries and 104 completed the online survey containing questionnaires (see Figure 5.1). Participants were recruited from six schools in Edinburgh (four schools) and East Lothian (two schools). Two of the participating schools were independent (one in Edinburgh and one in East Lothian) and the remaining four were state run.

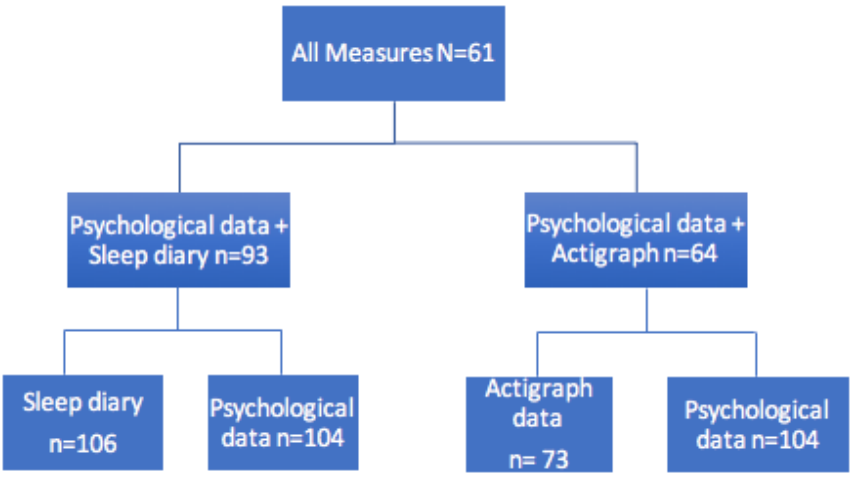


Figure 5.1: Participant Completion Rates for Study Components

Inclusion and exclusion Criteria

Participants were considered to be eligible for inclusion if they were between 13 and 18 years of age and self-identified to understand and speak English fluently. In line with the previous chapters, all adolescents regardless of mood symptomology were considered eligible. Individuals were required to provide informed consent and parental consent was obtained from participants under the age of 16. This study had no other exclusion criteria.

Measures

All basic information and measures of depression, anxiety, wellbeing, personality and emotional regulation that were administered online in relation to the previous study (Chapter Four) were utilised in this study with the addition of subjective and objective measures of sleep (see below). Briefly, Mood and Feelings Questionnaire (MFQ; Angold et al., 1995), Spence Children's Anxiety Scale (SCAS; Spence 1997), BBC-Well-being Scale (BBC; Kinderman, Schwannauer, Pontin and Tai, 2011) and the Eysenck Personality Questionnaire- Neuroticism subscale (Eysenck, Eysenck and Barrett, 1985) was employed here, as within the previous chapter.

In addition to those already mentioned, the following questionnaire measures were employed. All basic information and self-report measures were administered online hosted securely by Bristol Online Survey along with the questionnaire measures mentioned above:

Emotional Reactivity Scale (ERS, Nock, Wedig, Holmberg and Hooley, 2008)

The ERS is a 21-item self-report measure assessing individuals' experience of emotion reactivity. Each statement is responded to on a four-point scale (0= not at all like me, and 4= completely like me and comprises of three subscales assessing sensitivity, arousal and persistence. The ERS has demonstrated good internal consistency (α=.95) in this sample. The ERS was developed for use within adolescent samples.

Emotional Regulation Questionnaire for Children and Adolescents (ERQ-CA; Gullone and Taffe, 2011)

The ERQ-CA is a 10-item self-report measure of individuals' tendencies to employ reappraisal (six items) and suppression (four items) to regulate emotion, based on the adult scale developed by Gross and John (2003). Each item is measured on a seven-point Likert scale with one indicating 'strongly disagree' and seven indicating 'strongly agree' and subscales are scored as the mean of the items. This sample demonstrated good internal consistency (α=.76-.85)

Pubertal Development Scale (Petersen, Crockett, Richards and Boxer, 1987)

This measure was employed to assess development due to the implication of pubertal status within sleep preferences. This self-report measure of pubertal status demonstrated high internal consistency with (α =.77), and high correlation between self-report and physician ratings of puberty indicate the validity of this measure (Petersen et al., 1987). Participants were asked to respond to five questions (gender dependant) relating to physical development on a four-point scale (1=no development, 4=development completed).

Sleep

The Core Consensus Sleep Diary (CCSD; Carney et al. 2012)

The American Academy of Sleep Medicine developed the CCSD (Carney et al. 2012). This diary contains nine items considered to be the key parameters of sleep. The diary balances ease of use by the participant with the need for regular and reliable data gathering time points, and requires completion in the morning only. Questions related to the time of getting to bed; the time at which the individual attempted to fall asleep; sleep onset latency; number of

awakenings; duration of awakenings; time of final awakening; final rise time; perceived sleep quality, rated on a Likert scale (very poor, poor, fair, good, very good), as well as allowing for additional comments by the participant. To retain as high a sample size as possible, means of each component were employed for further analyses for all participants. Total sleep time as well as time in bed were calculated.

Actigraphy

In line with Ancoli-Israel et al.'s, (2003) guideline, Actigraphy was utilised in combination with sleep diaries to provide the most valid and reliable data. Philips Respironics Actiwatch 2 devices were employed and worn by participants for four weekday nights during typical school weeks. These Actiwatches recorded activity via an accelerometer and light exposure. Recorded data was scored using Philips Respironics Actiware Software. Standardised and default settings with medium sensitivity for this program were employed, in line with previous research (Meltzer, Walsh, Traylor and Westin, 2012). Software computed actograms of each participants movement and exposure to light. From this, software generated calculations of: sleep time, wake time, onset latency, wake after sleep onset (measure of sleep fragmentation), efficiency, time awake, percent awake, time asleep and percent asleep were generated.

Any effects of variability between individual devices was considered to be minimized as participation was arranged to maximise the numbers of participants that wore Actigraphs. As such, all ten of the devices were used equally, other than the final week of data collection where there were four participants. In this week, the devices to be used were selected at random. To increase reliability, participants were instructed to wear Actigraph watches for four school nights. A mean score over the four nights was calculated for each participant. Some data was missing due to participants having removed the Actigraph (further details below). In most cases this was due to individuals removing the Actigraph for activities such as showering and forgetting to put it back on. To retain as high a sample size as possible, means were employed for further analyses for all participants that had worn Actigraphs. One participants data was not downloadable due to a technical failure leaving the final sample size for this component at 73.

5.5 Protocol

Data for this study were collected alongside that of the previous study (Chapter Four). Briefly, participants completed online measures the same week as completing measures of sleep and

saliva measures of the previous study. After participants had been recruited, a participation schedule was devised in collaboration with the school contact. The aim here was to maximise the number of participants that could participate in the actigraphic component of the study, given constraints and schedules of the schools and the number of actigraphic devices (N=10). Schools that had greater flexibility allowed for involvement of pupils on consecutive weeks until all participants had completed in each component. Whereas, in schools with limited flexibility, often due to holidays, participation was limited to fewer weeks or single week participation and resulted in the random selection of participants to be involved in the actigraphic element.

Sleep diaries and wristwatches were given to and collected from participants alongside materials required for the previous study (Chapter Four). Typically, materials were distributed on the Monday of the participation week and collected on the Friday of the same week. Following the manufacturer's instructions, participants were instructed to wear the wristbands on their non-dominant wrist for the week of participation, removing only for activities involving water such as swimming and showering. Participants were instructed to answer the sleep diary questions every morning for one week. Participants were provided with a pack containing detailed instructions to refer to as well as, contact details if they had any queries. Data collected via actigraph data were downloaded over the weekend and stored digitally. For practical purposes of matching participants' data, actigraph devices were numbered (1-10) and the number of the actigraph provided to each participant was recorded using participant ID number.

At the end of data collection all participants were provided with a Debrief Sheet (see Appendix) which provided information regarding mental health support. Specifically, participants were advised that they may wish to speak to a trusted adult such as their parents, teachers or GP's if they had any concerns about their mood or mental health.

5.6 Statistical Analysis

Prior to analyses, distributions, skewness and kurtosis of raw data were examined. Continuous demographic variables and all non-time variables were within accepted limits of skewness and kurtosis, with the exception of puberty. Puberty was found to be leptokurtic (kurtosis=2.398). Further examination of this variable indicated that this may be driven by the higher proportion of females within the sample (n=81). Mean difference of puberty score between males and

females was examined. Independent samples t-test showed that females exhibited higher puberty scores than males ($t(102)=-2.69$, $p=0.008$). This indicates that the females within this sample had undergone greater pubertal development than males (mean difference of 1.71). This is not unexpected as developmental literature indicates that females begin puberty earlier than males (Rogol, Roemmich and Clark, 2002). As pubertal timing, age and gender are primarily employed as control variables and for the ease of interpretation, this variable has not been transformed. Furthermore, parametric statistical methods are considered robust enough to handle violations of assumptions.

Throughout analysis, time variables will be considered continuous, consistent with similar sleep research. All analyses involving time of day measures (e.g. bedtime) have been conducted to account for the 24-hour clock. Wrap-around functions were employed to ensure that times after midnight have been interpreted by analysis software as later than times before midnight. For example, a bedtime of 01:30 has been interpreted 3:30 later than 22:00.

A correction for multiple comparisons, (False Discovery Rate, calculated using R version 3.4.3 (2017-11-30; 'Kite-Eating Tree') has been applied to the significance statistics (p-values) within correlation tests.

Evidence of multicollinearity and autocorrelation has been considered in regression models. As the variables included initially are related to each other (e.g. percent sleep and percent wake), only one of the related variables was included within models. Within regression models measure of tolerance, Variance Inflation Factor and Durbin Watson fell within accepted limits (Field, 2009, p 297).

Hierarchical regression models, employing age, gender and puberty as the first step; neuroticism as the second step; and, sleep variables third. Sleep diary and actigraph variables were employed separately (i.e. in separate regression models), to avoid collinearity. Sleep diary variables consisted of the means of: sleep time, sleep onset latency, number of awakenings, wake after sleep onset, get up time, time in bed, total sleep time and sleep quality. Actigraph variables comprised means of; bedtime, wake time, sleep onset latency, number of awakenings, wake after sleep onset, percent sleep, time in bed, total sleep time and efficiency.

While controlling for age and gender, correlation analysis was conducted to examine relationships between measures of sleep and emotional regulation and reactivity

measures. Bootstrapping based on 1000 bootstrapped samples was employed. Analysis of sleep diary and actigraph measures were conducted separately to retain as much statistical power as possible.

5.7 Results

Gender differences

T-tests were employed in order to assess gender differences within the data. Females exhibited higher puberty scores than males ($t(102)=2.69$, $p=0.008$). This indicates that the females within this sample had undergone greater pubertal development than males (mean difference of 1.71). Females demonstrated significantly higher levels of anxiety ($t(59.64) =2.82$, $p=0.006$) and reduced levels of suppression as an emotional regulation strategy ($t(102=2.20$, $p=0.03$) compared to males. No gender differences were demonstrated in relation to sleep diary or actigraph variables (see Appendix).

Actigraph Compliance

Across the 73 participants, the number of valid nights that Actigraphic data was recorded ranged from 1 to 4, mean = 3.4 (SD= .76; mode=4); data were recorded for four nights for 53% of participants; three nights for 35.6% of participants; two nights for 8.2% of participants; and, one night for 2.7% of participants. When Actiwatches had been removed and not worn over night, participants most frequently reported forgetting to put them on after showering.

Sleep Diary Completion

Out of the 106 participants who returned completed sleep diaries, completion rates for all four nights was high, the mean number of completed nights was 3.8 (SD=0.4), ranging from 2-4 (mode=4); 86.8% of participants completed sleep diaries for four out of four nights; 11.3% of participants completed sleep diaries for three out of four nights; and. 1.9% of participants completed sleep diaries for two out of four nights. No participants only completed one night of the sleep diary. Figures 5.2 and 5.3 show the frequency of mean bed and wake times.

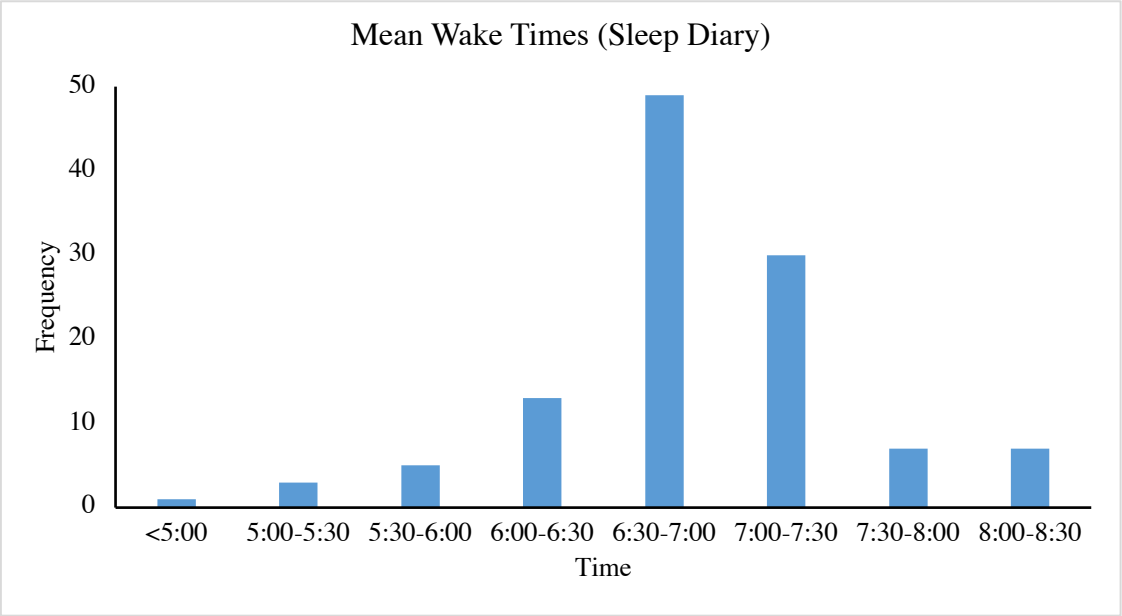


Figure 5.2: Frequency of Mean Wake Time (Sleep Diary)

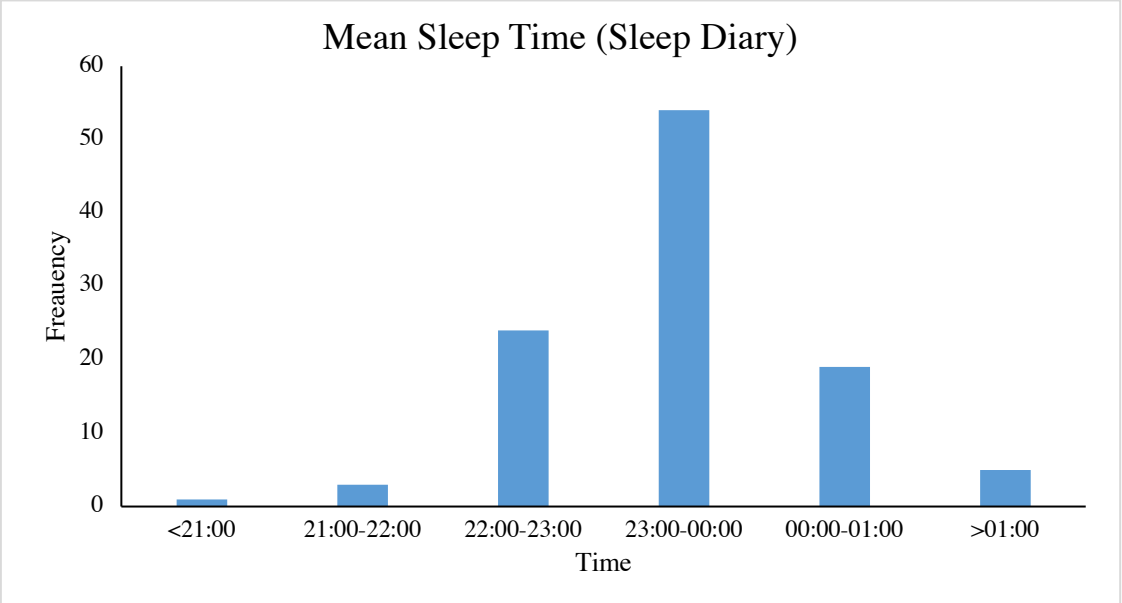


Figure 5.3: Frequency of Mean Bed Time (Sleep Diary)

Note: Time based on mean 'Try to sleep' time + mean sleep onset latency

Concurrence between subjective and objective sleep measures

Table 5.1: Sleep Diary and Actigraphy Concurrence

Measurement	Collection Method	Mean (S.D; N=69)	t(df), p
Bed Time	Diary	Into Bed: 20:56 Try to Sleep: 00:03	t(68)=7.03, p<0.001; t(68) =3.91, p<0.001
	Actigraph	22:04	
Total Sleep Time (minutes)	Diary	404.00 (160.32)	t(68)=3.94, p<0.001
	Actigraph	497.78 (104.20)	
Sleep Onset Latency (minutes)	Diary	21.68 (16.91)	t(68)=.48, p=0.633
	Actigraph	22.90 (20.94)	
Wake After Onset (minutes)	Diary	27.25 (21.04)	t(68)=-1.77, p=0.08
	Actigraph	33.02 (20.06)	
Awakenings (frequency)	Diary	0.72 (0.84)	t(68)=-17.42, p<0.001
	Actigraph	36.55 (17.26)	
Final Wake Time	Diary	Wake up: 06:46 Get up: 07:09	t(68)=-6.10, p<0.001; t(68)=4.85, p<0.001
	Actigraph	07:50am	

To compare similarity of measurements between self-report sleep diaries and objective actigraph measurements, comparisons between analogous measures have been conducted with correlation and t-tests (see Table 5.1).

Development and Sleep

Table 5.2: Pearson's Correlations of Sleep Diary Measurements and Development (FDR correction applied, n=92)

	In to Bed	Try to Sleep	Length to Sleep	Wake up	Get up	Awakenings (frequency)	Wake After Sleep Onset	Total Sleep Time	Quality Rating
Age	-0.03	-0.07	-0.16	0.16	.29*	.23*	-0.19	-0.04	-0.05
Sig. (2-tailed)	0.76	0.53	0.14	0.14	0.01	0.03	0.08	0.71	0.63
Puberty	-0.05	0.01	-.26*	0.10	0.18	0.01	-.34**	0.04	0.07
Sig. (2-tailed)	0.64	0.92	0.02	0.33	0.09	0.93	<0.01	0.73	0.52

Puberty and age demonstrated significant correlations with sleep diary (Table 5.2) and actigraph (Table 5.3) measured parameters of sleep. Pubertal development and age were significantly correlated (r=.47, p<0.001). Sleep diary measures of 'Get Up' time and frequency of awakenings were significantly correlated with age, while, length to sleep and wake after sleep onset were significantly correlated with pubertal development. Actigraph measures of Bed Time, Wake Time and Sleep Onset Latency were significantly correlated with age. Bed Time was significantly correlated with pubertal development.

Table 5.3: Actigraphy Measurements and Development (FDR correction applied, n=63)

	Bed Time	**Wake Time**	**Total Sleep Time**	**Onset Latency**	**Wake After Sleep Onset**	**Awakenings (frequency)**	**Efficiency**	**Percent Wake**	**Percent Sleep**
Age	.41**	-.36*	0.11	.28*	0.11	0.10	-0.16	0.06	-0.06
Sig. (2-tailed)	<0.01	0.01	0.39	0.03	0.38	0.47	0.22	0.66	0.66
Puberty	.35**	-0.03	0.11	-0.05	-0.03	-0.07	0.16	-0.10	0.10
Sig. (2-tailed)	0.01	0.83	0.41	0.72	0.83	0.60	0.22	0.45	0.45

Regression Analysis

See Table 5.4 for full results.

Overall regression models predicting MFQ were significant when including sleep diary and actigraph variables: r^2=0.55, F(11,80)=8.85, p<0.001 and r^2=0.44, F(13,49)=2.93, p<0.01, respectively.

Overall regression models predicting SCAS were significant when including sleep diary and actigraph variables: r^2=0.59, F(11,80)=10.66, p<0.001 and r^2=0.61, F(13,49)=5.93, p<0.001, respectively.

Table 5.4: Hierarchical Regression Models Using Sleep Diary and Actigraph Measures

Model		Model A (Sleep Diary, n=92)			Model B (Actigraph, n=63)		
DV	Statistic	Step 1 (Demographic)	Step 2 (Neuroticism)	Step 3 (Sleep Diary)	Step 1 (Demographic)	Step 2 (Neuroticism)	Step 3 (Actigraph)
MFQ	r^2	0.02	0.38	0.55	0.03	0.38	0.44
	r^2 change	0.02	0.36	0.17	0.03	0.36	0.05
	F (df1, df2)	(3,88) =0.49	(4,87) =13.02	(11,80) =8.85	(3,59) =0.58	(4,58) =9.03	(13,49) =2.93
	Significance Value (Model)	0.69	<0.001	<0.001	0.63	<0.001	<0.01
	Significance Value (F Change)	0.69	<0.001	<0.001	0.63	<0.001	0.86
SCAS	r^2	0.08	0.56	0.59	0.05	0.53	0.61
	r^2 change	0.08	0.48	0.04	0.54	0.47	0.08
	F (df1, df2)	(3,88) = 2.41	(4,87) =27.15	(11,80) =10.66	(3,59) =1.13	(4,58) =16.23	(13,49) =5.93
	Significance Value (Model)	0.07	<0.001	<0.001	0.35	<0.001	<0.001
	Significance Value (F Change)	0.07	<0.001	0.37	0.35	<0.001	0.34
BBC	r^2	0.06	0.37	0.45	0.01	0.40	0.50
	r^2 change	0.06	0.31	0.08	0.01	0.39	0.10
	F (df1, df2)	(3,88) =1.34	(4,87) =12.62	(11,80) =6.36	(3,59) =0.11	(4,58) =9.62	(13,49) =3.70
	Significance Value (Model)	0.15	<0.001	<0.001	0.95	<0.001	<0.001
	Significance Value (F Change)	0.14	<0.001	0.05	0.95	<0.001	0.42

Overall regression models predicting BBC were significant when including sleep diary and actigraph variables: r^2=0.45, F(11,80)=6.36, p<0.001 and r^2=0.50, F(13,49)=3.70, p<0.001, respectively.

In addition, the models employing sleep diary variables demonstrated a significant increase in r^2 from step two to step three for MFQ (Δr^2=0.17, ΔF(7,80)=4.42, p<0.001) and BBC (Δr^2=0.10, ΔF(7,80)=2.13, p=0.05), but not SCAS(Δr^2=0.04, ΔF(7,80)=1.10, p=0.37). This indicates that sleep diary variables accounted for a significant increase of variance above that of age, gender, pubertal status and neuroticism in predictions of MFQ and BBC but not in predictions of SCAS.

Individual sleep variables within regression models contributed significant unique variance (after controlling for all other variables). In predictions of MFQ, sleep diary 'try to sleep time' and sleep diary sleep onset latency demonstrated significant standardised beta-coefficients (sr^2=0.04, β=0.22, t(62)=2.70, p=0.008; and, sr^2=0.05, β=0.27, t(62)=3.07, p=0.003). No beta-coefficients were significant in predictions of SCAS. IN predictions of BBC, sleep onset latency demonstrated significant negatives standardised beta-coefficients (sr^2=0.04, β=(-)0.22, t(62)=-2.33, p=0.02). Full results available in Appendix.

The addition of actigraph variables at step three was not associated with a significant r^2 increase in predictions of MFQ (Δr^2=0.05, ΔF(9,49)=0.52, p=0.86), SCAS (Δr^2=0.08, ΔF(9,49)=1.17, p=0.34) or BBC (Δr^2=0.10, ΔF(9,49)=1.04, p=0.42).

In models using actigraph measures as predictors, no beta-coefficients of sleep measures were significant in predictions of MFQ. Actigraph measures of mean bed time and number of awakenings did demonstrate significant beta-coefficients (sr^2=0.04, β=0.26, t(62)=2.16, p=0.036; and sr^2=0.04, β=0.36, t(62)=2.14, p=0.04). Beta coefficients of mean wake time (actigraph measurement) were also, negatively, significant in predictions of BBC: sr^2=0.07, β=(-)0.38, t(62)=(-)2.68, p=0.01). Full results available in Appendix.

Correlational Relationships Between Sleep Parameters and Emotional Reactivity and Regulation

Results have been summarised in Table 5.5 and 5.6.

Table 5.5: Pearson's Bivariate Correlations with Sleep Diary Variables (n=92)

	GO TO BED	TRY TO SLEEP	LENGTH TO SLEEP	N. AWAKENINGS	LENGTH AWAKENINGS	WAKE UP	GET UP	QUALITY RATING
MFQ	0.13	0.30**	0.23*	-0.14	-0.01	0.15	0.30**	-0.34**
SCAS	0.12	0.19	0.05	-0.17	0.03	0.09	0.20	-0.26*
BBC	-0.05	-0.09	-0.11	0.11	0.03	-0.18	-0.20	0.25*
NEUROTICISM	-0.02	0.09	0.00	-0.10	0.06	0.05	0.15	-0.18
SENSITIVITY	0.01	0.03	-0.02	-0.23*	-0.03	0.00	0.04	-0.10
AROUSAL	0.05	0.13	0.00	-0.22*	-0.01	0.06	0.13	-0.17
PERSISTENCE	0.01	0.13	-0.03	-0.22*	-0.03	0.08	0.10	-0.22*
REAPPRAISAL	-0.18	-0.14	-0.07	0.02	0.04	-0.07	-0.06	0.00
SUPPRESSION	0.06	0.10	0.17	0.05	0.00	0.02	0.09	-0.20

**P<0.01, *P<0.05 Control variables: age, gender, puberty. Sleep variables based on averages from daily sleep diary entries.

Table 5.6: Pearson's Bivariate Correlations (r) with Actigraph Variables (n=63)

	BED TIME	SLEEP ONSET LATENCY	N. AWAKENINGS	WAKE AFTER SLEEP ONSET	TOTAL SLEEP TIME	%WAKE	WAKE UP	EFFICIENCY
MFQ	0.05	-0.04	-0.23	-0.24	0.10	-0.25	0.10	0.17
SCAS	0.08	-0.06	-0.03	-0.15	0.07	-0.19	-0.01	0.16
BBC	-0.12	0.03	0.11	0.12	0.03	0.10	-0.09	-0.07
NEUROTICISM	0.18	-0.07	-0.19	-0.19	0.02	-0.21	0.04	0.16
SENSITIVITY	-0.09	-0.12	-0.20	-0.20	0.12	-0.24	-0.03	0.21
AROUSAL	0.05	-0.02	-0.13	-0.21	0.00	-0.22	-0.07	0.12
PERSITENT	-0.01	-0.13	-0.17	-0.22	0.12	-0.29*	0.02	0.27*
REAPPRAISAL	0.14	0.08	0.05	0.05	-0.03	0.06	0.08	-0.05
SUPPRESSION	<0.01	-0.11	-0.23	-0.21	0.17	-0.23	0.20	0.19

**P<0.01, *P<0.05 Control variables: age, gender, puberty. Sleep variables based on averages from daily sleep diary entries.

The sensitivity subscale of the Emotional Reactivity Scale was significantly negatively correlated with the number of awakenings as measured with the sleep diary (r^2=0.04, p=0.05), and percent wake (r^2=0.06, p=0.05).

Arousal was significantly negatively correlated to sleep diary awakenings (r^2=0.05, p=0.04). Persistence was significantly positively correlate with Actigraph measures of efficiency (r^2=0.06, p=0.05) and negatively with percent wake (r^2=0.07, p=0.03); as well as, negatively correlated with sleep diary measures of awakenings (r^2=0.04, p=0.05) and positively correlated with the quality rating (r^2=0.05, p=0.03). Emotional regulation strategy measures were not significantly correlated with any sleep measures.

5.8 Discussion

Models predicting depression and anxiety symptoms and wellbeing each demonstrated overall significance when including either sleep diary or actigraph measures of sleep. The change in increase in r^2 following inclusion of sleep variables however, only demonstrated significance in the sleep diary model predicting depression and wellbeing, partially supporting our hypothesis. Contrary to the hypotheses, actigraph models did not offer additional predictive power of depression, anxiety or wellbeing symptoms. Nevertheless, in all cases a greater proportion of variance was explained when utilising sleep diary and actigraph variables as predictors. Like some previous findings, it may be that self-reported sleep behaviours are more salient predictors than objectively measured behaviours in relation to depressive symptomology.

The regression model of depression utilising sleep diary variables demonstrated significantly greater r^2. This supports previous literature demonstrating strong relationships between depression (diagnosis and symptom severity) and sleep behaviour.

Significant increase in explained variance of depression seems to be driven by sleep diary variables of 'try to sleep time' and sleep onset latency. Later try to sleep time and longer sleep onset latency were associated with increases of depression. Lower sleep onset latency was significantly related to higher wellbeing. This supports previous research indicating that increased time awake but in bed was associated with higher depression (e.g. Lovato and Grandsar, 2014). These findings are also in line with research demonstrating difficulties sleeping and longer night-time awakenings in individuals with depression. Objective measures of earlier wake time were related to higher wellbeing. This is aligned with previous research that found morning chronotypes as having improved health and wellbeing (e.g. Gulec et al., 2013).

In predictions of both anxiety and wellbeing, the models employing actigraph variables predicted a slightly greater proportion of variance than models employing sleep diaries. The converse was true for models of depression, where sleep diary variables predicted an increased proportion of variance. This indicates that self-reported sleep behaviour is a superior predictor of depression symptoms, while actigraph monitoring is marginally superior in predicting anxiety and wellbeing, within this sample.

These findings indicate a distinction between the presentation of depression and anxiety. An increased amount of variability of depression symptoms were accounted for by self-report measures whereas objectively captured measures were more salient predictors of anxiety. This is of interest considering the high comorbidity between anxiety and depression and that some theoretical explanations of these disorders indicate overlapping or indistinct underlying mechanisms contributing to a general risk for both anxiety and depression, such as neuroticism (Lahey, 2009). Furthermore, as sleep difficulties are symptomatic of both disorders, one may expect significant predictors to be the same.

Interestingly, no self-report measures were significantly related to anxiety. However, objective measures of sleep contributed a significant increase of explained variance. Higher anxiety was particularly related to later bed times and more awakenings during the night. Also, of note is that symptoms of anxiety include a greater number of physical and autonomic symptoms. For example, increased heart rate, sweating and nausea, which are not diagnostic components of depression (ICD-10, 1992). That objective differences in sleep are predictive of anxiety may reflect the physical and autonomic component present in anxiety, which is less pertinent to depression symptomology.

As predicted, and has been demonstrated in previous chapters, neuroticism was also significantly predictive of each MFQ, SCAS and BBC; accounting for approximately 30-40% of the variance of each of these psychological variables. This again highlights the salience of neuroticism in predicting psychological health. This finding is aligned with much research that implicates neuroticism in the development of psychological disorder.

Hirshkowitz, et al., (2015) recommended that adolescents achieve between 8 to 10 hours per night. Our results indicate that the mean self-reported total sleep time fell notably below this at only 6.5 hours (median=6.98; mode=7.25). The objectively measured sleep average fell just within the recommendation (8 hours 12 minutes). However, previous research has indicated that Actigraph measurement slightly over-estimates sleep time due to individuals having very low movement while in bed but awake (Weiss, Johnson, Berger and Redline, 2010; e.g. if watching television). As such, it may be prudent to consider both measures with caution and interpret the true value as falling somewhere between the two, in this case.

Consequently, these data support the assertion that adolescents do not achieve enough sleep, in line with previous findings. Furthermore, significant correlations between total sleep time

and puberty were slightly stronger than that of total sleep time and age. This may indicate that as individuals progress through development their sleep time becomes later, exhibiting the biologically driven chronotype shift. The present findings support previous research which indicated that the magnitude of the phase delay experienced by adolescents has been demonstrated to be unrelated to birth order, and school environment (proximity to older/younger students; Carskadon, Vieira and Acebo, 1993). This suggests that this behaviour is related to individuals' developmental age rather than familial or peer influence.

Late chronotype preference has itself been associated with deleterious health consequences. Arora and Taheri (2015) demonstrated that young adolescents with later chronotype were more likely to have unhealthy dietary habits and increased BMI. Urban, Magyarodi and Rigo, (2011) describe evening chronotype as 'health impairing' and employ structural equation modelling to demonstrate that later chronotype in adolescents increased the likelihood of smoking status and frequency, alcohol use and reduced physical activity. The relationships between adverse health and lifestyle effects and chronotype identifies this as an important marker of risk.

Prior research also indicated that this is in part due to early waking times as a result of school start conventions. As demonstrated in Figure 5.2, our sample demonstrated a high level of early start times with 59% waking between 5 and 6:30 am. An additional factor has previously been identified as the chronotype shift affecting adolescents, associated with later bed times and longer onset latencies. This is supported by the current findings which demonstrates that adolescents have a mean sleep onset latency of 20-25 minutes, which was also significantly related to age (actigraph measurement) and puberty (self-report measurement). As weekend sleep patterns were not measured within this sample, no conclusions can be made regarding free sleep periods in this sample. Reduced sleep may have significant long-term implications for adolescents. A recent large-scale study (N=14,894) by Smarr and Schirmer (2018) has demonstrated that 60% of students (age range 10-70) experience social jet lag and increases in the amplitude of this was strongly correlated with academic performance, particularly in late chronotypes. This suggests that not only is it difficult to adjust to socially imposed schedules, but that late chronotypes suffer more negative effects as a result compared to early chronotypes. However, the evidence did not indicate that late chronotpyes performed proportionately better in classes scheduled later in the day (Smarr and Schirmer, 2018). However, the study participants were mostly university students who face less consistency in classes which may contribute to a reduced ability to adjust to early schedules, resulting in

poorer performance across all times of the day due to social jet lag. A study conducted amongst Dutch high school pupils (age 11-18), where start times are consistent, demonstrated that lowest grades were obtained by those with very late chronotypes. However, the effect of chronotype on grades was no longer present in classers that occurred during the afternoon. It may be that late chronotypes are disadvantaged due to their biological predisposition. Conventional school starting times are more aligned to the preferences of early chronotypes and conflict with endogenous preference of late chronotypes. This may explain a significant amount of the difference of educational attainment between chronotypes. The constraint of school start may disproportionately impact late chronotypes resulting in this group underperforming in this context. Future research may address the impact of sleep duration and quality on academic performance and social impact as they are two high priority domains of functioning in this age group.

The salience of the subjective quality of sleep rating is also of interest. Only self-report sleep behaviours were predictive of the quality rating, no actigraph measures were predictive of the subjective rating. This suggests that subjective interpretation of sleep may be unrelated to observable behaviours. As discussed previously, it may be that these variables are mutually reinforcing; that negative mood negatively biases subjective experience of sleep quality. and/or that subjectively experienced poor-quality of sleep may bias subjectively experienced low mood. It may be that depressed individuals may report more difficulties resulting in an overestimation of their objective sleep difficulties or, healthy participants may underestimate any sleep difficulties. Further study may be necessary to disentangle this. Longitudinal approaches may be employed here to examine directionality of effect, as well as, experimentally manipulating mood state (e.g. employing negative mood induction). Similarly, studies examining sleep and mood variables in relation to measures of negative cognitive processing biases may aid understanding.

Emotional reactivity was significantly correlated with both sleep diary and Actigraph measurements, including: number of awakenings, quality, efficiency and percent wake. These correlations were all small but significant, indicating that emotional reactivity may be an important component in sleep quality and quantity, supporting previous research exploring the psychological conditions facilitating sleep. For example, Research indicates a relationship between maladaptive emotional regulation (rumination) and poor-quality sleep (Guastella and Moulds, 2007). Increased rumination has been associated with intrusive thoughts interfering with sleep. This has also been demonstrated through experimental manipulation.

Undergraduate students were assigned to rumination or distraction conditions, participants within the rumination condition demonstrated poorer sleep quality, particularly for those who demonstrated initial high trait rumination relative to low trait ruminators (Guastella and Moulds, 2007).

Further examination of these variables may improve understanding and provide potential avenues for interventions that may improve sleep quality and quantity, which may have knock-on consequences for health and wellbeing. These variables were not included in regression modelling due to issues of statistical power. Future work may address this by conducting a full mediation analysis of these variables.

Sleep difficulties may establishing non-stigmatised discussions of psychological health and wellbeing within a social context where mental health difficulties are particularly stigmatised. If sleep problems are less stigmatised than psychiatric symptoms, this may provide an alternative opportunity to identify and assess of psychological difficulties and risk thereby improving access to support. That sleep variables, particularly self-reported, were significantly related to depression, anxiety and wellbeing supports previous assertions of sleep as a transdiagnostic risk factor. The full predictive value of this could be further explored with longitudinal research approaches.

Limitations

There are several limitations to this study. Firstly, the nature of self-report measures which are associated with potential biases of reporting. For example, participants were asked to complete sleep diaries each morning following waking. However, there is no measure the extent to which participants complied with this. That there is less missing data for sleep diaries than actigraph may suggest that individuals completed the diary retrospectively rather than following waking as requested. Furthermore, the discrepancy between self-reported and objectively measured total sleep time demonstrated statistically significant difference and therefore both measures ought to be interpreted with caution. It is difficult to assess which measure has higher validity when considering that previous research indicates some measurement error within actigraph measurement and self-report is subject to reporter bias and researchers cannot be sure how closely participants followed the instructions to complete the diary first thing in the morning. If participants have retrospectively completed the diary at

the end of the week their reports would also be subject to recall bias in addition to self-report bias, such as social desirability bias.

In addition, predictions are based on regression analyses of cross-sectional data; therefore, this study is unsuited to determine causality and directionality of effects. Similarly, analysis has assumed linearity and as such, any non-linear effects have not been accounted for within this study.

A further issue may be the reliability of data captured by the actigraph and calculated by the actigraph software. A particular issue with actigraph measurement seems to be the overestimation of sleep time and lack of specificity in terms of distinguishing between inactivity in bed and sleep. Additionally, in a comparison study Meltzer et al, (2012) found adolescents to have the poorest specificity across actigraph devices and settings. However, the device chosen for this study captured ambient light exposure simultaneously in order to minimise this problem.

Additionally, although depression, anxiety and wellbeing symptoms have been measured as distinct from sleep measurements, there may be overlap in these measurements. Similarly, neuroticism has been measured as distinct from other psychological variables. Full mediation design would aid in detangling these variables, requiring a larger sample size.

As within the previous chapter, sample size and gender disparity are further limitations of this study. Due to missing data, the final sample size fell below the initial recruited target. This has important consequences for the power of statistical analysis. As such, there may be effects that have not been identified within this analysis due to lack of statistical power.

5.9 Conclusion

In conclusion, this study assessed sleep behaviour with both objective and subjective measurements to examine the relationship between psychological health and wellbeing, sleep and emotional reactivity and regulation. As sleep has been identified as an area of importance for various aspects of health, this is an important area to explore, particularly within the sensitive developmental window of adolescence. The identification of sleep difficulties may help detect risk of or presence of psychological disorder. This identification may be utilised in order to stratify adolescence into early intervention treatments, in the future. Self-reported

sleep behaviour demonstrated increased salience compared to objective measurements, this may provide an alternative route to identifying psychological problems within adolescents who may be reluctant to disclose mental health problems. Even though objective measurements did not account for any additional significant variance in psychological variables, it may be that such measurements are less useful in categorisation, or that objectively observable differences are not present in youth, as has been indicated by previous polysomnographic studies. Finally, the importance of neuroticism as a predictor of psychological health and wellbeing has been highlighted here again as this measurement was able to account for approximately 30-40% of unique variance of depression, anxiety and wellbeing symptoms.

Chapter Six - General Discussion

6.1 Introduction

This thesis has examined risk factors associated with depression and anxiety disorders. The areas are subject to limitations within previous literature, largely due to a scarcity of studies applied to the adolescent context. The design of each empirical chapter is similar in that the key outcome measurements are the same: depression (measured by the MFQ), anxiety (measured by SCAS) and wellbeing (measured by the BBC). Additionally, each chapter has employed regression analysis of cross-sectional data to predict each outcome variable beyond the variance that can be explained by neuroticism (as has been measured with the EPQ-N throughout). Neuroticism was identified as a key risk factor due to its heritable nature and (relative) stability over time. While some problems have been identified with the conceptualisations of neuroticism, which have been discussed in Chapter One, it remains a significant prospective predictor of future depression and anxiety. Furthermore, neuroticism has been considered a means of indexing general risk for internalising disorders. This chapter will summarise findings reported in Chapters Two to Five, discuss the implications of these findings and also consider the limitations of this research and areas that are worthy of future study.

6.2 Summary

A key strength of this thesis has been to consider the spectrum of mental health and do examine risk factors in relation to positive as well as negative affectivity. The positive side of the mental health spectrum is often overlooked within research, despite having been identified as of key importance in protecting against poor mental health, physical health and functioning more generally (e.g. social relationships, work and academic success etc., see Chapter One). Each chapter has considered predictions of both negative and positive symptoms (i.e. depression/anxiety and wellbeing). In order to fully capture a range of experiences of psychological health and wellbeing, participants were recruited from community settings.

To address the aims of the thesis and individual chapters, considerable amounts of data were been collected. Data collection was completed in two rounds, both of which employed substantial protocols. The design and implementation of these protocols were particularly labour intensive. Recruitment of adolescents through schools is challenging, requiring considerable time and effort in communicating with schools, individual teachers, pupils and

parents. The data collection for this study was also particularly time consuming. Data collection for Chapters Two and Three took approximately 45-60 minutes per participant. Data collection for Chapters Four and Five required less one-to-one time with individual participants but required frequent visits to school sites, more complex organisation in order to collect samples and equipment, and to ensure samples were stored appropriately as soon as possible. Participants generally enjoyed participating in this research, particularly the physiological components involved in Chapters Four and Five. Both participants and schools were been interested in the research and dissemination of results. Building strong relationships with contacts within schools was a key priority in order to maximise the success of these studies, and any potential future projects that may take place within local schools. Coordinating and conducting recruitment and data collection comprised a significant portion of this body of work.

Throughout this thesis, neuroticism has predicted a large proportion of depression, anxiety and wellbeing measures (between 30-55%). Importantly, the contribution of neuroticism has remained consistent between both sets of data collected. Contributions of neuroticism were highest for anxiety and lowest for wellbeing. Similar to neuroticism, attachment has been much examined within psychological research and has been robustly associated with psychological disorders. Chapter Three identified that parental attachment was more salient compared to peer attachment in relation to depression and wellbeing. Peer attachment was identified as important for anxiety; however, its inclusion did not significantly increase predictions of anxiety. Building on the regression models developed in Chapter Two, the inclusion of parental attachment variables significantly increased predictions of depression and wellbeing. It was identified that there was some overlap between contributions of neuroticism and attachment although both explained unique variance.

Chapter Two and Three demonstrated that cognitive styles and cognitive biases contributed unique variance to predictions of depression, anxiety and wellbeing. These predictors remained significant in Chapter Three when attachment variables were included in regression models. The overall the proportion of variance of depression, anxiety and wellbeing was increased in Chapter Three compared to models from Chapter Two. Biases of interpretation and self-referential recall were particularly salient predictors. Salience of different realms of cognitive bias differentially predicted depression and anxiety. Results indicate that symptoms of depression and anxiety are not associated with a globally negative bias, but a bias of information relating to the self. The proportion of variance explained by cognitive variables

was highest for wellbeing and lowest for anxiety. This may identify cognitive biases as a target for intervention to promote wellbeing which may protect against disorder onset.

Cognitive biases also explained a significant proportion of variance over and above both neuroticism and attachment. Cognitive contributions may be more easily modified (e.g. CBT) compared to biologically driven or early childhood factors. One potential confound is that neuroticism may interact with cognitive biases to impact upon depression or depressive risk. Results of this thesis identify that relationships between cognitive bias and depression were stronger than those between cognitive bias and neuroticism. Additionally, neuroticism and cognitive biases explained unique variance within predictions, indicating that they may play distinct roles. However, full mediation analysis could be employed to elucidate this more fully. The biases that most strongly drove relationships/ predictions in this thesis were self-referential memory biases and interpretation bias. This provides support to Beck's theory of depression involving negative schema relating to the self, the world and others. So, challenging these cognitive biases may improve depression symptoms or interrupt the cycle of negativity thereby challenging depressive states.

The focus of Chapters Four and Five turned to potential physiological markers of disorder or disorder risk. The hypothesised role of the HPA axis was investigated through measures of evening and morning cortisol concentrations, cortisol awakening response and daily cortisol production. Additionally, a measure of longitudinal (previous three months) exposure to cortisol was investigated. These measures were employed as predictors in regression models predicting depression, anxiety and wellbeing, but no significant increase of variance was demonstrated. Nevertheless, significant standardised beta-coefficients indicated that higher bedtime and lower waking cortisol concentration were associated with an increase in depression. Similarly, higher waking cortisol concentrations were associated with higher wellbeing. These findings again distinguish depression from anxiety. Additionally, emotional reactivity and regulation measures were found to be associated with cortisol concentration and reactivity, respectively. This supports the hypothesis of a relationship between psychological emotional regulation and physiological stress response. Consequently, intervention to modify emotional reactivity and regulation style may have a positive impact on cortisol exposure, which is proposed to affect disease risk.

Previous research has not provided clarity of relationships between HPA axis activity and disorders. These findings do not provide strong support for a relationship between the stress-

response system and psychological health. Some weak relationships were however, demonstrated between cortisol concentrations and depression and wellbeing. As such, this area is worthy of further study within a larger sample in order to increase the statistical power to detect effects. It may also be that these relationships are more complex than has been demonstrated within the analysis. For example, there may be a distinction in the role of the stress-response system in relation to specific subtypes or symptoms of depression (such as anhedonia, increased versus decreased sleep and increased versus decreased appetite). For example, the current results indicate that higher evening and lower morning cortisol (contributing to lower variation) may be related to depression. This component is also linked to arousal and as such may be interpreted as being important to symptoms of depression related to fatigue, lack of motivation, and slowness.

Chapter Five focused on sleep behaviour which comprises a distinct risk factor for emotional disorders but has also been related to cortisol concentrations (due to circadian influences) and emotional processing. This chapter aimed to improve examination of this risk factor by combining self-report sleep diary and the objective actigraphic measurement of sleep. Only self-report sleep variables were found to significantly increase the explained variance of depression (but not anxiety or wellbeing), from that which could be explained by neuroticism. Objective measurements failed to significantly predict any of the key outcome variables. However, standardised beta-coefficients did demonstrate that objective measures of later sleep time and increased sleep onset latency did significantly impact depression. This supports previous literature which has identified self-report measures as superior predictors and may indicate that there are some subtle objective differences that are related to depressive aetiology. However further examination of these variables is necessary to fully address this. Results from this chapter also supported previous findings indicating that adolescents face below optimal sleep duration, which has the potential to have deleterious consequences, both in terms of physical and psychological health as well as, social and academic functioning.

6.3 Limitations

Each Chapter has addressed limitations specific to the empirical studies described. However, there are several limitations impacting the research described within this thesis more generally. Firstly, each study has relied upon cross-sectional design. Consequently, this research has been unable to assess directionality of effects. Although each study has focused on predicting depression, anxiety and wellbeing, neither the directionality nor causal impact of these can be determined. Therefore, due to lack of temporal information, results can only support

associations between constructs. Re-evaluating the components examined within this thesis within longitudinal designs is an avenue for future research which would help identify causal mechanisms.

The self-report nature of personality, cognitive, emotional, psychological and sleep variables are subject to biases. Although the measures employed have demonstrated reliability (within this study) and validity (in previous research), they remain subject to potential biases. For example, self-report measures are likely subject to desirability bias, variation of the introspective ability of individuals, individuals' understanding or interpretation of questions, and responder bias; steps were taken to minimise the likelihood of such biases, for example all such measures were completed independently and anonymously. Additionally, the voluntary nature of the recruited sample is a potential bias. It is possible that individuals who volunteered to participate in this research demonstrate particular characteristics that increase their propensity for participating. For example, individuals who have experience of depression or anxiety may be more inclined to participate in a research study investigating mental health. To minimise this, the study was offered to all pupils within the participating schools (or all those that the school facilitated participation of i.e. certain classes or year groups that were identified by the school). These biases may limit the generalisability of findings to all adolescents.

The studies described within this thesis recruited participants within Edinburgh and Lothians. Edinburgh is a city that has a particularly high number of independent schools. In order to recruit as large a sample as possible, both state and independent schools were contacted in regard to participation. Two (different) independent schools participated in each round of data collection. Approximately 30-40% of the sample was recruited from independent schools. This figure is higher than the number of children that attend independent secondary schools within Edinburgh (~25%, Denholm, 2017). As such there is a potential socio-economic bias within the recruited samples that may limit the generalisability of findings. Nevertheless, state schools that participated included those whose catchment area covers some of the 5% most deprived areas within Scotland (two schools, based on the Scottish Index of Multiple Deprivation, 2016). This may go some way to balance the socioeconomic demographics of the samples. However, unfortunately no socioeconomic information was gathered as part of this study.

A further limitation throughout these studies has been limited power due to sample size. Recruiting adolescents is notoriously difficult; initial backwards-elimination regressions were underpowered due to sample size. As such, there is an increased risk of Type II error here, variables excluded based on removal criteria may demonstrate undetected effects. Although target sample sizes were recruited, the number of participants with complete data fell below the target sample size. This limited the statistical power of analysis and the complexity of analysis that could be conducted. Measures have been employed to reduce the impact of this and have been described within each chapter (for example, p-value adjustments for multiple comparisons, bootstrapping and reducing the number of variables included within regression models). Larger sample sizes would allow for more complex models and full mediation analysis of related variables, an avenue for future research.

There are also potential drawbacks of the analysis techniques employed within this thesis. Chapters Two and Three employed backwards elimination regression, a method of stepwise regression. Stepwise methods were selected to aid the identification of most salient predictors given the limited power to include all predictors. Limitations of stepwise methods were considered prior to analysis and attempts were made to minimise these. A backwards method was chosen over a forward method due to the increase of suppressor effects associated with forwards methods (Field, 2009). Due to suppressor effects forwards methods are associated with a higher risk of Type II error. As such, backwards methods are generally preferred over forwards methods. Consequently, backwards methods were employed within this thesis. However, stepwise methods in general have disadvantages. For example, they run a risk of over and under-fitting. Furthermore, predictors are selected by software based on statistical criteria rather than theoretical importance. Consequently, theoretically important variables may have been excluded from final models. For example, in Chapter Three this method selected a peer attachment variable and eliminated a parental attachment variable in predictions of anxiety. Parental attachment was selected in predictions of depression and wellbeing and is theoretically considered of greater importance than peer attachment. Therefore, this model may be limited due to inclusion of peer rather than parental attachment.

A further criticism of stepwise methods is that predictors are conditionally selected based on other predictors that have already been selected (Thompson, 1995). Therefore the 'best' predictors vary depending upon predictors that are already in (or left in; depending on a backwards or forwards method) the model. Variables are included/excluded based on their ability to explain unique variance of the dependent variable further than that of the first

selected variable (e.g. neuroticism in chapter three). Thompson (1995) proposes a more optimal strategy would be to consider the contribution of all possible subsets of variables to find the combination that best fit. With the methods employed within Chapters Two and Three it is possible that there is a different combination of the variables selected that explain a larger amount of variance of the dependent variables. Due the problems with stepwise methods, they have been employed within this research as a means of selecting predictors, not hypothesis testing. Forced entry methods are considered more reliable than stepwise methods and have been proposed as the only appropriate method for theory testing (Studenmund and Cassidy, 1987). As such, forced entry method was employed for hypothesis testing.

6.4 Future directions

Overall, the results of this thesis indicate that physiological components were certainly less strongly predictive of depression, anxiety and wellbeing compared to psychological measures. The importance of the personality contribution of neuroticism as well as attachments has been highlighted throughout this thesis. This indicates that a significant proportion of risk can be predicted by factors that are partly genetically driven and related to early life experience. As such, modification of these variables may be particularly difficult. Nevertheless, these may be useful means of identifying individuals at risk, particularly as these seem to remain fairly stable over time. Therefore, continuing research and creativity within this area is of high importance.

While neuroticism is considered stable over time, previous research does not consider it 'fixed' until adulthood. Consequently, modification of neuroticism within childhood and adolescence may be more successful than within adulthood when personality is considered to be more fixed. The shared variance of neuroticism and attachment demonstrated within Chapter Two may indicate shared underlying mechanisms or that these factors impact upon each other (i.e. neuroticism may contribute to maladaptive attachment, and/or vice versa). As such addressing these factors could be of key importance. As well as potentially reducing risk for psychological disorders, improving attachment security may improve interpersonal relationships - important for overall wellbeing.

Cognitive mechanisms were highlighted as predictive of depression, anxiety and wellbeing. A focus of future research may include identifying additional cognitive mechanisms that contribute to disorder aetiology, for example, reward processing. Research identifying cognitive mechanisms that are specific to adolescent processing may improve the success of

cognitive interventions within this age group. The salience of self-referential information may also indicate that identifying biases relevant to specific individuals may also help inform disorder onset.

Future research may focus on examining risk factors simultaneously. For example, developing a means of measuring neuroticism, attachments, cognitive biases and sleep over time (such as through a mobile application) would allow recruitment of large numbers which may highlight those who are potentially vulnerable. Then, identification of specific risk factors could inform tailored treatment. Alongside this cortisol data could be collected and employed as an objective measure to examine any improvement over time alongside self-report measures. Developing research that can be delivered through mobile technology would also have benefits of allowing for frequent sampling, sampling at random time points and for superior ecological validity. Applications which could set reminders or deliver notifications may also improve attrition rates inherent to research. Adopting technology within research may also be very amenable to individuals (particularly young people), as mobile technology is so integrated into everyday life, and allow for research to reach a wider group of people.

Conducting a large longitudinal study employing significant predictors identified within this thesis alongside other known predictors (e.g. family history, childhood adversity, polygenetic risk scores and neuroimaging) would be advantageous in furthering understanding and the relative contributions of these factors. This would have potential implications for clinical practice in terms of identifying those at risk, and identify disorder subtypes that may result in individuals experiencing a more chronic and recurrent form of illness.

The neural plasticity associated with adolescents, due to ongoing development, identifies this period as a potential target for intervention to improve wellbeing, and minimise risk for depression and anxiety. Substantial physical and neurological development occurs at different rates between male and female adolescents. This, as well as the disparity in prevalence of psychological disorders between genders may support the examination of risk and disorder aetiology in relation to genders independently, rather than considering males and females as one group. While adolescents are particularly vulnerable to experiencing depression, this is the age where girls become particularly at risk and it is during adolescence that the gender difference, which prevails through adulthood, originates. Gjerde and Block (1996), demonstrated significant gender differences in predictors of onset of depression in adolescence; for girls, maternal depression during childhood was strongly related to onset,

while onset in boys was related to deficits in early supportive care. In general, mental illness is considered to rise in adolescence (Kessler et al., 2005), Costello, Mustillo and Erkanli et al., (2003) demonstrated that transition to adolescence was marked by increase of depression and social phobia in girls that was not demonstrated in boys. This replicates Cohen et al., (1993), who found that girls exhibited a sharp increase of depression instances at this age, until a peak at around age 14. Whereas, for boys, instances of depression were at their lowest point upon beginning adolescence and rose gradually until age 20. Future work to identify the specific biological mechanisms by analysing males and females separately and longitudinally and may provide greater insight to causal mechanisms.

This thesis supports the involvement of increased wakefulness during time in bed is associated with depression. Future research examining the role of sleep in larger, longitudinal and prospective studies would clarify this association and further knowledge. Preventative intervention focusing on improving sleep quality may minimise onset of depression, studies examining the minimisation of wakefulness in bed may identify this as a protective factor in the onset of disorders. Additionally, it has been demonstrated that in general adolescents achieve below optimal sleep durations. Furthermore, this work supports the notion that adolescents in general are achieving below optimal sleep. As this has been identified as a risk factor for poor outcomes across various realms of functioning, generalised interventions to adolescents promoting positive sleep practices may improve health, wellbeing and functioning of adolescents.

6.5 Conclusion

Adolescence is a risk period for the onset of psychological disorders. However, limited research focuses on this age group. Treatments for depression have demonstrated lower efficacy in adolescents than within adults, for whom they are designed. Psychological disorders presenting within this age group are marked by symptoms that differ from symptoms occurring within adult patients. Additionally, adolescence is a period of substantial developmental change, potentially impacting the mechanisms involved in the development of disorders. Consequently, research addressing risk factors within this unique period is a priority within mental health research, and has been the focus of this thesis. This thesis has presented empirical research that has identified strong links between symptoms of depression and anxiety with lower neuroticism, poorer attachment quality, stressful-life events, negative cognitive biases of self-referential recall and interpretation, emotional reactivity and emotional regulation strategies. Self-reported sleep behaviour has been strongly linked to depression symptoms. Links between HPA-axis activity and objective measures of sleep have also been implicated in depressive, but not anxious symptoms. Factors that have been identified as important within this thesis have demonstrated significance above and beyond the role of neuroticism. A second focus of this thesis has been to examine the role of risk factors in wellbeing. Throughout this thesis, wellbeing has been identified as distinct to that of depression and anxiety. Additionally, factors that have been predictive of depression have also been implicated in wellbeing, supporting the theoretical understanding of depression and psychological wellbeing existing on a continuum. This may also support previous research identifying wellbeing as a protective factor against the onset of disorders. Further research, building on these findings, may establish more efficacious treatment for disorder as well as early intervention treatments to promote wellbeing and minimise risk of disorder development.

References

Adam, E.K., (2006) Transactions among adolescent trait and state emotion and diurnal and momentary cortisol activity in naturalistic settings, *Psychoneuroendocrinology, 31(5), 664-79*

Adam E.K., and Kumari, M., (2009) Assessing salivary cortisol in large-scale, empidemiological research, *Psychoneuroendocrinology*, 34, 1423-1436

Adam, E.K., Quinn, M.E., Tavernier, R., McQuillan, M.T., Dahlke, K.A., Gilbert, K.E., (2017) Diurnal cortisol slopes and mental and physical health outcomes: a systematic review and meta-analysis, *Psychoneuroendocrinology,* 83, 25-41

Adam E. K., Doane, L.D., Zinbarg, R.E., Mineka, S., Craske M.G., and Griffith, J.W., (2010) Prospective prediction of major depressive disorder from cortisol awakening responses in adolescence, *Psychoneuroendocrinology,* 35(6), 921-931

Adam, E.K., Vrshek-Schallhorn, S., Kendall, A.D., Mineka, S., Zinbarg, R.E., and Craske, M.G., (2014), Prospective associations between the cortisol awakening response and rifst onsets of anxiety disorders over a six-year follow up – 2013 Curt Richter Award Winner, *Psychoneuroendocrinology*, 44,44-59

Agerup, T., Lydersen, S., Wallander, J., Sund, A.M., (2015) Associations between parental attachment and course of depression between adolescence and young adulthood, *Child Psychiatry and Human Development*, 46(4), 632-42

Ainsworth, M. D. S., Blehar, M. C., Waters, E., and Wall, S. (1978). Patterns of attachment: A psychological study of the strange situation. Oxford, England: Lawrence Erlbaum.

Ainsworth, M. D. S., Bell, S. M., and Stayton, D. J. (1971) Individual differences in strange- situation behavior of one-year-olds. In H. R. Schaffer (Ed.) *The origins of human social relations*. London and New York: Academic Press. Pp. 17-58.

Alfano, C.A., Zakem, A.H., Costa, N.M., Taylor, L.K., and Weems, C.F., (2009) Sleep problems and their relation to cognitive factors, anxiety, and depressive symptoms in children and adolescents, *Depression and Anxiety,* 26(6), 503-12

Allen, J.P., Porter, M., and McFarland, C., (2007) The relation of attachment security to adolescents' parental and peer relationships, depression, and externalizing behaviour, *Child Development,* 78(4), 1222-1239

Alloy, L.B., Ahrens, A.H., 1987. Depression and pessimism for the future: biased use of statistically relevant information in predictions for self versus others. *J. Pers. Soc. Psychol.* 52 (2), 366–378.

Alvaro, P.K., Roberts, R.M., and Harris, J.K., (2013) A systematic review assessing bidirectionality between sleep disturbances, anxiety and depression, *Sleep*, 36(7), 1059:1068

Ancoli-Israel, S., Cole, R., Alessi, C., Chambers, M., Moorcroft, W., and Pollak, C.P., (2003) The role of actigraphy in the study of sleep and circadian rhythms, *Sleep*, 26(3), 342-392

Angold, A., Costello, E.J., Messer, S.C., Pickles, A., Winder, F., Silver, D., 1995. Development of a short questionnaire for use in epidemiological studies of depression in children and adolescents. *Int. J. Methods Psychiatr. Res.* 5, 237–249.

Angst, J., and Merikangas, K.R., and Preisig, M., (1997) Subthreshold syndromes of depression and anxiety in the community, *Journal of Clinical Psychiatry*, 58 Suppl 8, 6-10

American Psychiatric Association (1987) Diagnostic and Statistical Manual of Mental Disorders (3rd ed., rev.; DSM-III-R), Washington, DC

American Psychiatric Association (2013) The Diagnostic and Statistical Manual of Mental Disorders (5th ed., DSM-5), Washington, DC

Armsden, G. C., & Greenberg, M. T. (1987). The Inventory of Parent and Peer Attachment: Individual differences and their relationship to psychological well-being in adolescence. *Journal of Youth and Adolescence, 16*(5), 427-454.

Avenevoli, S., Swendsen, J., He, J.P., Burstein, M., Merikangas, K.R., 2015. Major de- pression in the national Comorbidity Survey- Adolescent Supplement: prevalence, correlates, and treatment. *J. Am. Acad. Child Adolesc. Psychiatry* 54 (1), 37–44 e2.

Avery, L.J.R., LeBourgeois, M.K., Gupta, T., and Mittal, V.A., (2015) Actigraphic measured sleep disturbance predicts positive symptoms in adolescents at ultra-high-risk for psychosis: a longitudinal study, *Schizophrenia Research* 164(1-3), 15-20

Barclay, N.L., Eley, T.C., Parsons, M.J., Willis, T.A., and Gregory, A.M., (2013) Monozygotic twin differences in non-shared environmental factors associated with chronotype, *Journal of Biological Rhythms,* 28(1), 51-61

Barrett, P.M., and Holmes, J., (2001) Attachment relationships as predictors of cognitive interpretation and response bias in late adolescence, *Journal of Child and Family Studies*, 10(1), 51-64

Bartels, M., Cacioppo, J.T., van Bejisterveldt, T.C., Boomsma, D.I., 2013. Exploring the association between well-being and psychopathology in adolescents. *Behav. Genet.* 43 (3), 177–190.
Bartholomew, K., and Horowitz, L.M., (1991) Attachment styles among young adults: a test of a four-category model, *Journal of Personal and Social Psychology,* 61(2) 226-44
Baum, K.T., Desai, A., Field, J., Miller, L.E., Rausch, J., and Beebe, D.W., (2014) Sleep restriction worsens mood and emotion regulation in adolescents, *Journal of Child Psychology and Psychiatry,* 55(2), 180-190
Beck, A.T., (1967) Depression- Clinical experimental and theoretical aspects, New York: Harper and Row
Beck, A.T., (2008) The evolution of the cognitive model of depression and its neurobiological correlates, *Am. J. Psychiatry,* 165(8), 969-77
Benca, R.M., Obermeyer, W.H., Thisted R.A., and Gillin, J.C., (1992) Sleep and psychiatric disorders. A meta-analysis, *Arch. Gen. Pscyhiatry*, 49(8), 651-668
Benjamini, Y., Hochberg, Y., 1995. Controlling the false discovery rate: a practical and powerful approach to multiple testing. *J. Royal Stat. Soc. Ser. B* (Methodol.) 57 (1), 289–300.
Bertocci, M.A., Dahl, R.E., Williamson, D.E., Iosif, A.M., Birmaher, B., and Axelson, D., and Ryan, N.D., (2005) *J. Am. Acad. Child Adolescent Psychiatry,* 44(11), 1158-66
Binz, T.M., Braun, U., Baumgartner M.R., and Kraemer T., (2016) Development of an LC-MS/MS method for the determination of endogenous cortisol in hair using (13)C3-labeled cortisol as a surrogate analyte, *J. Chromatography, B. Analytical Technology in Biomedical Life Sciences*, 1033-1034
Borel, B., (2015) If it feels like stress is killing you that's because it might be, *Popular Science: Health https://www.popsci.com/chronic-stress-it-could-be-killing-you*
Bouhuys, A.L., Geers, E., Gordijn, M.C.M., 1999. Depressed patients' perceptions of facial emotions in depressed and remitted states are associated with relapse: a longitudinal study. J. Nerv. Ment. Dis. 187, 595–602.
Bourke, C., Douglas, K., Porter, R., 2010. Processing of facial emotion expression in major depression: a review. Aust. N. Z. J. Psychiatry 44 (8), 681–696.
Bowen, R., Baetz, M., Leuschen, C., & Kalynchuk, L. E. (2011). Predictors of suicidal thoughts: Mood instability versus neuroticism. *Personality and Individual Differences, 51*, 1034–1038.
Bowlby, J., (1969/1982) Attachment and loss: Vol. 1. Attachment. 2. New York: Basic Books; 1969/1982.
Bowlby, J., (1973) Attachment and loss: Vol. 2. Separation: Anxiety and anger. New York: Basic Books; 1973.
Bowlby, J. ,(1980), Attachment and loss: Vol. 3. Sadness and depression. 2. New York: Basic Books; 1980.
Brennan, K.A., Clark, C.L., and Shaver, P.R., (1998) Self-report measurement of adult romantic attachment: an integrative overview. In Simpson, J.A., and Rholes, W.S., (Eds.), Attachment theory and close relationships (pp.46-47) New York: Guilford Press
Burke, H.M., Davis, M.C., Otte, C., and Mohr, D.C., (2005) Depression and cortisol responses to psychological stress: a meta-analysis, *Psychoneuroendocrinology*, 30(9), 846-56
Buysse, D.J., Reynolds, C.F., Monk, T.H., Berman, S.R., Kupfer, D.J., (1989) Pittsburgh Sleep Quality Index: a new instrument for psychiatric practice and research, *Psychiatric Research*, 28(2), 193-213
Cacioppo, J.T., Bernston, G.G., Malarkey, W.B., Kiecold-Glaser, J.K., Sheridan, J.F., Poehlmann, K.M., Burleson, M.H., Ernst, J.M., Hawkely, L.C., and Glasser, R., (1998) Autonomic, neuroendocrine, and immune responses to psychological stress: The reactivity hypothesis, *Annals of the New York Academy of Science,* 840, 664-673
Carney, C.E., Buysse, D.J., Ancoli-Israel, S., Edinger, J.D., Krystal, A.D., LIchstein, K.L., Morin, C.M., (2012) The consensus sleep diary: standardizing prospective sleep self-monitoring, *Sleep*, 35(2), 287-302
Carskadon, M., (2002) Adolescent sleep patterns: Biological, social and psychological influences, Cambridge, UK: Cambridge University Press
Carskadon, M., Vieira C., and Acebo, C., (1993) Association between puberty and delayed phase preference, *Sleep*, 16(3) 258-62
Casey, B.J., Jones, R.M., Hare, T.A., 2008. The Adolescent Brain. Annals of the New York Academy of Sciences 1124, 111–126.
Cellini, N., Duggan, K.A., and Sarlo, M., (2017) Perceived sleep quality: the interplay of neuroticism, affect and arousal, *Sleep Health*, 3(3) 184-189
Chan, S., Goodwin, G., Harmer, C., 2007. Highly neurotic never-depressed students have negative biases in information processing. Psycholog. Med. 37 (9), 1281–1292.
Chida, Y., and Steptoe, A., (2008) Positive psychological well-being and mortality: a quantitative review of prospective observational studies, *Psychosomatic Medicine,* 70(7) 741-56

Choudhury, S., Blakemore, S.J., Charman, T., (2006) Social cognitive development during adolescence, *Social Cognitive and Affective Neuroscience*, 1(3) 165-74
Chrousos, G.P., and Gold, P.W., (1992) The concepts of stress and stress system disorders. Overview of physical and behavioural homeostasis, *JAMA*, 267(9), 1244-52
Chung, S., Son, G.H., and Kim, K., (2011) Circadian rhythm of adrenal glucocorticoid: its regulation and clinical implications, *Biochimicia et Biophysica Acta (BBA)-Molecular Basis of Disease*, 1812(5), 581-591
Clark, L.A., Watson, D., and Mineka, S., (1994) Temperament, personality, and the mood and anxiety disorders, *Journal of Abnormal Psychology*, 103(1) 103-16
Clarke, G., and Harvey, A.G., (2012). The Complex Role of Sleep in Adolescent Depression, *Child and Adolescent Psychiatric Clinics of North America*, 21(2) 385-400
Cohen, S., Janicki-Deverts, D., and Miller, G.E., (2007) Psychological stress and disease, *JAMA*, 298(14) 1685-7
Collins, N.L., and Read, S.J., (1994) Cognitive representations of attachment: The structure and function of working models. In K. Bartholomew & D. Perlman (Eds.), *Advances in personal relationships, Vol. 5. Attachment processes in adulthood* (pp. 53-90). London, England: Jessica Kingsley Publishers.
Connolly, S.L., Abramson, L.Y., Alloy, L.B., 2016. Information processing biases con- currently and prospectively predict depressive symptoms in adolescents: evidence from a self-referent encoding task. Cognit. Emot. 30 (3), 550–560.
Costello, E.J., Erkanli, A., and Angold, A., (2006) Is there an epidemic of child or adolescent depression? *Journal of Child Psychology and Psychiatry*, 47(12)
Costello, E.J., Mustillo, S., and Erkanli , A., Keeler, G., and Angold, A., (2003) Prevalence and development of psychiatric disorders in childhood and adolescence, *Archives of General Psychiatry*, 60(8) 837-44
Cowen, P.J., (2002) Cortisol serotonin and depression: all stressed out? *British Journal of Psychiatry*, 180, 99-100
Crawford, T. N., Livesley, W. J., Jang, K. L., Shaver, P. R., Cohen, P., and Ganiban, J. (2007). Insecure attachment and personality disorder: A twin study of adults. *European Journal of Personality, 21*(2), 191-208.
Crawford, T.N., Shaver, P.R., and Goldsmith, H.H., (2007) How affect regulation moderates the association between anxious attachment and neuroticism, *Attachment and Human Development*, 9(2), 95-109
Crowell, J.A., and Waters, E., (1994) Bowlby's Theory Grown Up: the role of attachment in adult love relationships, *Psychological Inquiry*, 5(1)
Crowley, S.J., Acebo, C., and Carskadon, M.A., (2007) Sleep, circadian rhythms, and delayed phase in adolescence, *Sleep Medicine*, 8, 602-612
Cicchetti, D., Cummings, E. M., Greenberg, M. T., & Marvin, R. S. (1990). An organizational perspective on attachment beyond infancy: Implications for theory, measurement, and research. In M. T. Greenberg, D. Cicchetti, & E. M. Cummings (Eds.), *The John D. and Catherine T. MacArthur Foundation series on mental health and development. Attachment in the preschool years: Theory, research, and intervention* (pp. 3-49). Chicago, IL, US: University of Chicago Press.
Cummings, C. M., Caporino, N. E., & Kendall, P. C. (2013). Comorbidity of anxiety and depression in children and adolescents: 20 years after. *Psychological bulletin*, *140*(3), 816-45.
Dahl, R.E., (2004) Adolescent brain development: a period of vulnerabilities and opportunities. Keynote Address, *Annals of the New York Academy of Science*, 1021, 1-22
Dahl, R.E., and Lewin, D.S., (2002) Pathways to adolescent health sleep regulation and behaviour, *Journal of Adolescent Health*, 6, 175-84
Dahl, R.E., (2007) Sleep and the Developing Brain, *Sleep*, 30(9) 1079-1080
Danielsson, N. S., Jansson-Fröjmark, M., Linton, S. J., Jutengren, G., and Stattin, H. (2010). Neuroticism and sleep-onset: What is the long-term connection? *Personality and Individual Differences, 48*(4), 463-468.
Danner, F., and Phillips, B., (2008) Adolescent sleep, school start times, and teen motor vehicle crashes, *Journal of Clinical Sleep Medicine*, 4(6) 533-535
Daviss, W.B., Birmaher, B., Melhem, N.A., Axelson, D.A., Michaels, S.M., Brent, D.A., 2006. Criterion validity of the Mood and Feelings Questionnaire for depressive episodes in clinic and non-clinic subjects. J. Child Psychol. Psychiatry 47 (9), 927–934.
Dedovic, K., and Ngiam, J., (2015) The cortisol awakening response and major depression: examining the evidence, *Neuropsychiatric Distress Treatment*, 11, 1181-1189

Derksen, S., and Keselman, H.J., (1992) Backward, forward and stepwise automated subset selection algorithms: frequency of obtaining authentic and noise variables, *British Journal of Mathematical and Statistical Psychology*, 45(2)
Drevets, W.C., (2001) Neuroimaging and neuropathological studies of depression: implications for the cognitive-emotional features of mood disorders, *Current Opinions in Neurobiology*, 11(2), 240-90
Druss, B.G., Hwang, I., Petukhova, M., Sampson, N.A., Wang, P.S., Kessler, R., (2009) *Molecular Psychiatry*, 14(7), 728-37
Duckworth, A.L., Steen, T.A., and Seligman, M.E., (2005) Positive psychology in clinical practice, *Annual Review of Clinical Psychology*, 1, 629-51
Dunn, V., Goodyer, I.M., 2006. Longitudinal investigation into child-and adolescence- onset depression: psychiatric outcome in early adulthood. Br. J. Psychiatry 188 (3), 216–222.
Everaert, J., Duyck, W., Koster, E.H.W., 2015. Emotionally biased cognitive processes: The weakest link predicts prospective changes in depressive symptom severity. *PLoS ONE* 10 (5), e0124457.
Eysenck, S.B.G., Eysenck, H.J., Barrett, P., 1985. A revised version of the psychoticism scale. Pers. Individ. Differ. 6, 21–29.
Feldman, R., (2017) The Neurobiology of Human Attachments, *Trends in Cognitive Sciences*, 21(2) 80-99
Fergusson, D.M., Boden J.M., and Horwood, L.J., (2007) Recurrence of major depression in adolescence and early adulthood, and later mental health, eduational and economic outcomes, *Brittish Journal of Psychiatry,* 191, 335-42
Fergusson, D.M., Woodward, L.J., and Horwood, L.J., (2000) Risk factors and life processes associated with the onset of suicidal behaviour during adolescence and early adulthood *Psychological Medicine*, 30(1) 23-39
Field, T., (1996), Attachment and separation in young children, *Annual Review of Psychology*, 47, 541-61
Field, A., 2009. Discovering Statistics Using SPSS, 3rd Ed. Sage Publications Ltd, London.
Fisak, B.J. Jr., Richard, D., and Mann, A., (2011) The prevention of child and adolescent anxiety: a meta-analytic review, *Prevention Science*, 12(3); 255-68
Fredriksen, K., Rhodes, J., Reddy, R., and Way, N., (2004) Sleepless in Chigaco: tracking the effects of adolescent sleep loss during the middle school years, *Child Development*, 75(1) 84-95
Foulkes, L., and Blakemore, S-J., (2018) Studying individual differences in human adolescent brain development, *Nature Neuroscience*, 21:3 (315-323)
Ganzola, R., Nickson, T., Bastin, M.E., Giles, S., Macdonald, A., … Duchesne, S., (2017) Longitudinal differences in white matter integrity in youth at high familial risk for bipolar disorder, *Bipolar Disorders,* 19(3)
Garcia-Diaz, E., Quintana-Gallego, E., Ruiz, A., Carmona-Bernal, C., and Benhamou-Botebol, (2007) Respiratory polygraphy with actigraphy in the diagnosis of sleep apnea-hyponea syndrome, *Chest,* 131(3) 725-732
Gau, S.F., (2000) Neuroticism and sleep-related problems in adolescence, *Sleep*, 23(4) 495-502
Giedd, J.N., Blumenthal, J., Jeffries, N.O., Castellanos, F.X., Liu, H., Zijdenbos, A., Paus, T., Evans, A.C., Rapport, J.L., (1999) Brain development during childhood and adolescence: a longitudinal MRI study, *Nature Neuroscience*, 2(10) 861-3
Gjerde, P.F., and Block, J., (1996) A developmental perspective on depressive symptoms in adolescence: gender differences in autocentric-allocentric models of impulse regulation, n Cicchetti, D., and Toth, S.L., (Eds.), *Rochester symposium on developmental psychopathology, Vol. 7. Adolescence: Opportunities and challenges* (pp. 167-196). Rochester, NY, US: University of Rochester Press.
Gold, P.W., and Chrousos, G.P., (2002) Organization of the stress system and its dysregulation in melancholic and atypical depression: high vs low CRH/NE states, *Molecular Psychiatry,* 7(3) 254-75
Goodyer, I.M., Reynolds, S., Barrett, B., Byford, S., Dubicka, B., Hill, J., Fonagy, P., (2017). Cognitive behavioural therapy and short term psychoanalytical intervention in adolescents with unipolar major depressive disorder (IMPACT): a multicentre, pragmatic, observer-blind randomised controlled superiority trial. *The Lancet Psychiatry* 4(2
Goodyer, I.M., Herbert, J., Tamplin, A., Altham, P.M.E., (2000) Recent life events, cortisol, dehdroepiandrosterone and the onset of major depression in high-risk adolescents, *The British Journal of Psychiatry*, 177(6) 499-504
Goodyer, I.M., Herbert, J., Tamplin, A., Altham, P.M., (2000) First-episode major depression in adolescents. Affective, cognitive and endocrine characteristics of risk status and predictors of onset, *British Journal of Psychiatry*, 176, 142-9

Goodyer, I.M., Herbert, J., Tamplin, A., Secher, S.M., and Pearson, J., (1997) Short-term outcome of major depression: II. Life events, family dysfunction, and friendship difficulties as predictors of persistent disorder, *Journal of the American Academy of Child and Adolecent Psychiatry*, 36(4) 474-80

Gordon, J., and Platt, S., (2017) Going to be…all right? – A report on the mental health of young people in Scotland, commissioned by SAMH https://www.samh.org.uk/documents/Going_to_Be_All_Right_Jacki_Gordon_Report_2017.pdf

Gregory, A.M., and Eley, T.C., (2005) Sleep problems, anxiety and cognitive style in school-aged children, *Infant and Child Development*, 14(5)

Gregory, A.M., and Sadeh, A., (2012) Sleep, emotional and behavioural difficulties in children and adolescents, *Sleep Medicine Review*, 16(2) 129-36

Gregory, A.M., and Sadeh, A., (2016) Annual Research Review: Sleep problems in childhood psychiatric disorders –a review of the latest science, *Journal of Child Psychology and Psychiatry*, 57(3) 296-317

Griffin, A., (2017) Adolescent neurological development and implications for health and well-being, *Healthcare (Basel)*, 5(4). Pii: E62

Griffith, J.W., Kleim, B., Sumner, J.A., Ehlers, A., 2012. The factor structure of the autobiographical memory test in recent trauma survivors. Psychol. Assess. 24 (3),

Guastella, A.J., and Moulds, M.L., (2007) The impact of rumination on sleep quality following a stressful life event. *Personality and Individual Differences, 42*(6), 1151-1162.

Gulec, M., Selvi, Y., Boysan, M., Aydin, A., Oral, E., and Aydin, E.F., (2013) Chronotype effects on general well-being and psychopathology levels in healthy young adults, *Biological Rhythm Research,* 44(3), 457-468

Gulliver, A., Griffiths, K.M., and Christensen, H., (2010) Perceived barriers and facilitators to mental health help-seeking in young people: a systematic review, *BMC Psychiatry*, 10:113

Gullone, E., and Robinson, K., (2005) The Inventory of Parent and Peer Attachment – Revised, (IPPA-R) for children: a psychometric investigation, *Clinical Psychology and Psychotherapy*, 12(1)

Gullone, E., and Taffe, J., (2011) The emotion regulation questionnaire for children and adolescents (ERQ-CA): a psychometric evaluation, *Psychological Assessment*, 24(2), 409-417

Gunaydin, G., Selcuk, E., and Ong, A.D., (2016) Trait reappraisal predicts affective reactivity to daily positive and negative events, *Frontiers in Psychology*, 7, 1000

Hagenauer, M.H., Perryman, J.I., Lee, T.M., and Carskadon, M.A., (2009) Adolescent changes in the homeostatic and circadian regulation of sleep, *Developmental Neuroscience*, 31(4) 276-84

Harvey, C., and Peirson, S., (2015), What Makes You Tick? Oxford Sparks, Bringing Oxford Science to Life, https://www.oxfordsparks.ox.ac.uk/what-makes-you-tick

Harvey, A.G., Murray, G., Chandler, R.A., Soehner, A., (2011) Sleep disturbance as transdiagnostic: consideration of neurobiological mechanisms, *Clinical Psychology Review*, 31(2), 225-235

Hauner, K. K. Y., Adam, E. K., Mineka, S., Doane, L. D., DeSantis, A. S., Zinbarg, R., … Griffith, J. W. (2008). Neuroticism and Introversion are Associated with Salivary Cortisol Patterns in Adolescents. *Psychoneuroendocrinology*, *33*(10), 1344–1356.

Hazan, C., and Shaver, P.R., (1987) Romantic love conceptualised as an attachment process, *Journal of Personality and Social Psychology*, 52(3), 511-524

Hazan, C., Shaver, P.R., (1990) Love and work: an attachment-theoretical perspective, *Journal of Personality and Social Psychology*, 59(2), 270-280

Hazan, C., and Zeifman, D., (1990) Pair bonds as attachments: evaluating the evidence In J. Cassidy & P. R. Shaver (Eds.), Handbook of attachment: Theory, research, and clinical applications. New York: Guilford Press

Helhammer, J., Fries, E., Schweisthal, O.W., Schlotzz, W., Stone, A.A., and Hagemann, D., (2007) Several daily measurements are necessary to reliably assess the cortisol rise after awakening: state- and trait components, *Psychoneuroendocrinology*, 32(1), 80-6

Hebert, J., (2013) Cortisol and depression: three questions for psychiatry, *Psychological Medicine*, 43(3), 449-69

Hettema, J.M., Neale, M.C., Myers, J.M., Prescott, C.A., Kendler, K.S., 2006. A population-based twin study of the relationship between neuroticism and internalizing disorders, *The American Journal of Psychiatry*, 163(5), 857-64

Hettema, J., (2008) What is the genetic relationship between anxiety and depression?, *Am J Med Genet C Semin Med Genet*, May 15; 148C(2):140-6

Hirschfield, R.M., (2001) Comorbidity of Major Depression and Anxiety Disorders: recognition and management in primary care, *Primary Care Companion Journal of Clinical Psychiatry*, 3(6) 244-254

Hirshkowitz, M., Whiton, K., Albert, S.M., Alessi, C., Bruni, O., … Hillard, P.J.A., (2015) National Sleep Foundation's Sleep time duration recommendations: methodology and results summary, *Sleep Health*, 1(1)40-43

HM Treasury (2003) Every Child Matters, ISBN 0101586027,

Hoyt, L.T., Craske, M.G., Mineka, S., and Adam, E.K., (2015) Positive and negative affect and arousal: cross-sectional and longitudinal associations with adolescent cortisol diurnal rhythms, *Psychosomatic Medicine*, 77(4), 392-401

International Classification of Mental and Behavioural Disorders: Clinical Descriptions and Diagnostic Guidelines, 10th Revision (ICD-10), (1992), Geneva, World Health Organisation

Irons, C., and Gilbert, P., (2005) Evolved mechanisms in adolescent anxiety and depression symptoms: the role of the attachment and social rank systems, *Journal of Adolescents*, 28(3), 325-41

Ivaneko, A., Crabtree, V.M., and Gozal, D., (2005) Sleep and depression in children and adolescents, *Sleep Medicine Review*, 9(2) 115-29

Jaworska, N., and MacQueen, G., (2015) Adolescence as a unique developmental period, *Journal of Psychiatry and Neuroscience*, 40(5), 291-3

Jenni, O.G., and LeBourgeois, M.K., (2010) Understanding sleep-wake behaviour and sleep disorders in children: the value of a model, *Current Opinion in Psychiatry*, 19(3), 282-287

Jones-Mason, K., Allen, I. E., Bush, N., & Hamilton, S. (2016). Epigenetic marks as the link between environment and development: examination of the associations between attachment, socioeconomic status, and methylation of the SLC6A4 gene. *Brain and behavior*, *6*(7), e00480. doi:10.1002/brb3.480

Joormann, J., Gilbert, K., Gotlib, I.H., 2010. Emotion identification in girls at high risk for depression. *Journal of Child Psychology and Psychiatry, and Allied Disciplines,* 51 (5),

Kalmbach, D.A., Schneider, L.D., Cheung, J., Bertrand, S.J., Kariharan, T., Pack, A.I., and Gehrman, P.R., (2017) Genetic basis of chronotype in humans: insights from three landmark GWAS, *Sleep, 40(2)*

Karlen, J., Ludvigsson, J., Frostell, A., Theodorsson, E., Farejo, T., (2011) Cortisol in hair measured in young adults – a biomarker of major life stressors? *BMC Clinical Pathology*, 11-12

Kaufman, J., Martin, A., King, R.A., and Charney, D., (2001) Are child-, adolescent-, and adult-onset depression one and the same disorder? *Biological Psychiatry*, 49(12) 980-1001

Keenan, K., Hipwell, A., Babinski, D., Bortner, J., Henneberger, A., Hinze, A., … Sapotichne, B. (2013). Examining the developmental interface of cortisol and depression symptoms in young adolescent girls. *Psychoneuroendocrinology*, *38*(10), 2291–2299

Kelvin, R.G., Goodyer, I.M., Teasdale, J.D., Brechin, D., 1999. Latent negative self-schema and high emotionality in well adolescents at risk for psychopathology. J. Child Psychol. Psychiatry 40 (6), 959–968

Kendler, K.S., Neale, M.C., Kessler, R.C., Health, A.C., Eaves, L.J., (1993) A longitudinal twin study of personality and major depression in women, *Archives of General Psychiatry*, 50(11), 853-62

Kendler, K.S., Kuhn, J., and Prescott, C.A., (2004) The interrelationship of neuroticism, sex and stressful life events in the prediction of episodes of major depression, *The American Journal of Psychiatry*, 161(4), 631-6

Kendler, K.S., Gatz, M., Gardner, C.O., and Pederse, N.L., (2006) Personality and major depression: a Swedish longitudinal population-based twin study, *Archives of General Psychiatry*, 63(10), 1113-20

Kercher, A.J., Rapee, R.M., and Schniering, C.A., (2009) Neuroticism, life events and negative thoughts in the development of depression in adolescent girls, *Journal of Abnormal Child Psychology*, 37(7), 903-15

Kessler, R.C., Berglund, P., Delmer, O., Jin, R., Merikangas, K.R., and Walters, E.E., (2005) Lifetime prevalence and age-of-onset distributions of DSM-IV disorders in the National Comorbidity Survey Replication, *Archives of General Psychiatry*, 62(6), 593-602

Kessler, R.C., Berglund, P., Delmer, O., Jin, R., Merikangas, K.R., Walters, E.E., and Wang, P.S., (2003) The epidemiology of major depressive disorder: results from the National Comorbidity Survey Replication (NCS-R), *JAMA*, 289(23), 3095-105

Khan, A.A., Jacobson, K.C., Gardner, C.O., Prescott, C.A., and Kendler, K.S., (2005) Personality and comorbidity of common psychiatric disorders, *British Journal of Psychiatry*, 186, 190-196

Kinderman, P., and Cooke, A., (2017) Responses to the publication of the American Psychiatric Association's DSM 5, *Journal of Humanistic Psychology*

Kinderman, P., Schwannauer, M., Pontin, E., Tai, S., 2011. The development and validation of a general measure of well-being: the BBC well-being scale. *Qual. Life Res.* 20 (7), 1035–1042.

Kirschbaum, C., and Hellhammer, D.H., (1989) Salivary cortisol in psychobiological research: an overview, *Neuropsychobiology*, 22(3), 150-69

Klein, D.N., Kotov, R., Bufferd, S.J., 2011. Personality and depression: Explanatory models and review of the evidence. *Annu. Rev. Clin. Psychol.* 7, 269–295.
Kornreich, C., and Philippot, P., (2006) Dysfunctions of Facial emotional recognition in adult neuropsychiatric disorders: influence on interpersonal difficulties, *Psychologica Belgica*, 46(1-2), 79-98
Kotov, R., Gamez, W., Schmidt, F., Watson, D., 2010. Linking "big" personality traits to anxiety, depressive, and substance use disorders: a meta-analysis. *Psychol. Bull.* 136 (5), 768–821.
Kudielka, B.M., and Kirschbaum, C., (2005) Sex differences in HPA azis responses to stress: a review, *Biological Psychology*, 69(1), 113-32
Kuyken, W., Watkins, E., Holden, E., and Cook, W., (2006) Rumination in adolescents at risk for depression, *Journal of Affective Disorders,* 96(1-2), 39-47
La Greca, A.M., and Harrison, H.M., (2010) Adolescent peer relations, friendships, and romantic relationships: do they predict social anxiety and depression? *Journal of Clinical Child Adolescent Psychology*, 34(1), 49-61
Lahey, B.B., 2009. Public health significance of neuroticism. *Am. Psychol.* 64 (4), 241–256
Laible, D.J., Carlo, G., and Raffaelli, M., (2000) The differential relations of parent and peer attachment to adolescent adjustment, *Journal of Youth and Adolescence*, 29(1), 45-59
Lawrence, D., Johnson, S., Hafekost, J., de Haan, K.B., Sawyer, M., Ainley, J., and Zubrick, S.R., (2015) The mental health of children and adolescents: report on the second Australian child and adolescent survey of mental health and wellbeing, Australian Council for Educational Research, Department of Health, Canberra
Le Blanc, M., Beaulieu-Bonneau, S., Merette, C., Savard, J., Ivers, H., Morin, C.M., (2007) Psychological and health-related quality of life factors associated with insomnia in a population-based sample, *Journal of Psychosomatic Research*, 63(2), 157-66
Lee, A., Hankin, B.L., 2009. Insecure attachment, dysfunctional attitudes, and low self- esteem predicting prospective symptoms of depression and anxiety during adolescence. *J. Clin. Child Adolescent Psychol.* 28 (2), 219–231.
Lee, D. Y., Kim, E., & Choi, M. H. (2015). Technical and clinical aspects of cortisol as a biochemical marker of chronic stress. *BMB Reports*, *48*(4), 209–216.
Lemoine, P., Gilleminault, C., and Alvarez, E., (2007) Improvement in subjective sleep in major depressive disorder with a novel antidepressant, agomelatine: randomized, double-blind comparison with venlafaxine, *Journal of Clinical Psychiatry*, 68(11), 1723-32
Lemola, S., Ledermann T., and Friedman, E.M., (2013) Variability of sleep duration is related to subjective sleep quality and subjective well-being: an actigraphic study, *PloS One,* 8(8)e
Levendosky, A.A., Huth-Bocks, A., and Semel, M.A., (2010) Adolescent peer relationships ane mental health functioning in families with domestic violence, *Journal of Clinical Child and Adolescent Psychology*, 31(2), 206-218
Lewinsohn, P. M., Rohde, P., & Seeley, J. R. (1998). Major depressive disorder in older adolescents: Prevalence, risk factors, and clinical implications. Clinical Psychology Review, 18, 765–794.
Lichstein, K.L., Stone, J.C, Donaldson, J., Nau, S.D., Soeffing, J.P., Murray, D., Lester, K.W., Aguillard, R.N., (2006) Actigraphy validation with insomnia, *Sleep,* 29(2), 232-9
Lovato, N., and Gradisar, M., (2014) A meta-analysis and model of the relationship between sleep and depression in adolescents: recommendations for future research and clinical practice, *Sleep Medicine Review,* 18(6), 521-9
Lucinao, M., Hagenaars, S.P., Davies, G., Hill, W.D., Clarke, T-K., Shirali, M., Deary, I.J., 2018. Association analysis in over 329,000 individuals identifies 116 independent variants influencing neuroticism. *Nature Genetics* 50, 6–11.
Malouff, J.M., Thorsteinsson, E.B., Schutte, N.S., 2005. The relationship between the five- factor model of personality and symptoms of clinical disorders: a meta-analysis. J. *Psychopathological Behavioural Assessment* 27 (2), 101–114.
Mannie, Z.N., Harmer, C.J., and Cowen, P.J., (2007) Increased waking salivary cortisol levels in young people at familial risk of depression, *American Journal of Psychiatry*, 164(4), 617-21
March, J., Silva, S., Petrycki, S., Curry, J., Wells, K., Fairbank, J., Burns, B., Domino, M., McNulty, S., Viteiello, B., Severe, J, Treatment for Adolescents with Depression Study (TADS) Team, Fluoxetine, cognitive-behavioural therapy, and their combination for adolescents with depression: treatment for adolescents with depression study (TADS) randomised controlled trial, *JAMA*, 292(7), 807-20
Marco, E.M., Marci, S., Laviola, G., 2011. Critical age windows for neurodevelopmental psychiatric disorders: evidence from animal models. *Neurotoxicology Research* 19 (2), 286–307.

Marcia, J.E., (1980) Identity in adolescence in Adelson, J., (Ed.*) Handbook of Adolescent Psychology*, Wiley (1980)
Margolese, S.K., Markiewicz, D., and Doyl, A.B., (2005) Attachment to parents, best friend, and romantic partner: predicting different pathways to depression in adolescence, *Journal of Youth and Adolescence*, 34, 673
Marsh, A.A., Ambady, N., and Kleck, R.E., (2005) The effects of fear and anger facial expressions on approach and avoidance-related behaviours, *Emotion,* 5(1), 119-24
Mathew, S.J., Coplan, J.D., Goetz, R.R., Feder, A., Greenwald, S., Dahl, D.E., Ryan, N.D., Mann J.J., and Weissmann, M.M., (2003) Differentiating depressed adolescent 24h cortisol secretion in light of their clinical outcome, *Neuropsychopharmacology*, 28(7), 1336-43
Mazurka, R., Wynne-Edwards, K.E., and Harkness, K.L., (2016), Stressful life events prior to depression onset and the cortisol response to stress in youth with first onset versus recurrent depression, *Journal of Abnormal Child Psychology*, 44(6), 1173-84
McEwan, B.S., (1998) Stress adaptation, and disease. Allostasis and allostatic load, *Annals of the New York Academy of Science*, 840, 33-44
McEwen, B.S., and Stellar, E., (1993) Stress and the individual. Mechanisms leading to disease, *Archives of International Medicine*, 153(18), 2093-101
McEwen, B.S., (2000) The neurobiology of stress: from serendipity to clinical relevance, *Brain Research*, 886(1-2), 172-189
McLaughlin., K.A., and Hatzenbuehler, M.L., (2009) Stressful life events, anxiety sensitivity and internalising symptoms in adolescence, *Journal of Abnormal Psychology,* 118(3), 659-69
McLaughlin, K.A., Nole-Hoeksema, S., 2011. Rumination as a transdiagnostic factor in depression and anxiety, *Behavioural Research and Therapy*, 49(3), 186-93
Meltzer, L.J., Walsh, C.M., Traylor, J., and Westin, A.M., (2012) Direct comparison of two new actigraphs and polysomnography in children and adolescents, *Sleep*, 35(1), 159-66
Merikangas, K.R., He, J.P., Burstein, M., Swanson, S.A., Avenevoli, S., Cui, L., Swendsen, (2010) Lifetime prevalence of mental disorders in U.S. adolescents: results from the National Comorbidity Survey Replication – Adolescent Supplement (NCS-A), *Journal of the American Academy of Child and Adolescent Psychiatry*, 49(10), 980-90
Merikangas, K.R., Zhang, H., Avenevoli, S., Acharyya, S., Neuenschwander, M., Angst, J., and Zurich Cohort Study, (2003) Longitudinal trajectories of depression and anxiety in a prospective community study: the Zurich Cohort Study, *Archives of General Psychiatry*, 60(10), 993-1000
Micco, J.A., Henin, A., Hirshfeld-Becker, D.R., 2014. Efficacy of interpretation bias modification in depressed adolescents and young adults. *Cognition Therapy and Research* 38 (2), 89–102.
Miguel-Hidalgo, J.J., (2013) Brain structural and functional changes in adolescents with psychiatric disorders, *International Journal of Adolescent Medical Health*, 25(3), 245-56
Mor, N., Winquist, J., 2002. Self-focused attention and negative affect: a meta-analysis. *Psychological Bulletin*. 128 (4), 638–662.
Moussavi, S., Chatterji, S., Verdes, E., Tandon, A., Patel, V., and Ustun, B., (2007) Depression, chronic diseases and decrements in health: results from the World Health Surveys, *Lancet*, 370(9590):851-8
MQ, (2016) Young People's Mental Health, https://www.mqmentalhealth.org/mental- health/children-and-young-people, 09/10/17.
Muris, P., van Brakel, A. M. L., Arntz, A., & Schouten, E. (2011). Behavioral Inhibition as a Risk Factor for the Development of Childhood Anxiety Disorders: A Longitudinal Study. *Journal of Child and Family Studies*, *20*(2), 157–170
Muris, P., Merckelbach, H., Ollendick, T., King, N., and Bogie, N., (2002) Three traditional and three new childhood anxiety questionnaires: their reliability and validity in a normal adolescent sample, *Behavioural Research and Therapy*, 40(7), 753-72
National Health Service Scotland Health Scotland (2016), Good mental health for all, ISBN: 978-1-84485-592-6,
National Collaborating Centre for Mental Health (UK; 2005) *Depression in Children and Young People: Identification and Management in Primary, Community and Secondary Care*. *Leicester* (UK): British Psychological Society; 2005. (NICE Clinical Guidelines, No. 28.) Available from: https://www.ncbi.nlm.nih.gov/books/NBK56425/
Navrady, L.B., Ritchie, S.J., Chan, S.W.Y., Kerr, D.M., Adams, M.J., Hawkins, E.H., McIntosh, A.M., 2017. Intelligence and neuroticism in relation to depression and psychological distress: evidence from two large population cohorts. *Eur. Psychiatry* 43, 58–65.

Niedenthal, P.M., Brauer, M., Robin L., and Innes-Ker, A.H., (2002) Adult attachment and the perception of facial expressions of emotion, *Journal of Personality and Social Psychology*, 83(2), 419-33

Nock, M.K., Wedig, M.M., Holmberg, E.B., and Hooley, J.M., (2008) The emotion reactivity scale: development, evaluation and relation to self-injurious thoughts and behaviours, *Behavioural Therapy* 39(2), 107-16

Noftle, E. E., & Shaver, P. R. (2006). Attachment dimensions and the big five personality traits: Associations and comparative ability to predict relationship quality. *Journal of Research in Personality, 40*(2), 179-208

Nolen-Hoeksema, S., 2000. The role of rumination in depressive disorders and mixed anxiety/depressive symptoms. *J. Abnorm. Psychol.* 109 (3), 504–511.

Novati A, Roman V, Cetin T, Hagewoud R, den Boer JA, Luiten PG, et al. (2008) Chronically restricted sleep leads to depression-like changes in neurotransmitter receptor sensitivity and neuroendocrine stress reactivity in rats. *Sleep* 31:1579–1585.

Oakley, N.R., (1997) Validation with polysomnography of the Sleep-watch sleep/ wake scoring algorithm used by the Actiwatch activity monitoring system. Technical Report to Mini Mitter Co., Inc., 1997

Ochsner, K. N., & Gross, J. J. (2008). Cognitive Emotion Regulation: Insights from Social Cognitive and Affective Neuroscience. *Current Directions in Psychological Science*, *17*(2), 153–158.

Orchard, F., Pass, L., Reynolds, S., (2016) 'It was all my fault'; Negative interpretation bias in depressed adolescents. *Journal Abnormal Child Psychology* 44 (5), 991–998.

Ormel, J., Jeronimus, B. F., Kotov, R., Riese, H., Bos, E. H., Hankin, B., … Oldehinkel, A. J. (2013). Neuroticism and Common Mental Disorders: Meaning and Utility of a Complex Relationship. *Clinical Psychology Review*, *33*(5), 686–697

Ormel, J., Oldehinkel, A. J., & Vollebergh, W. (2004). Vulnerability before, during, and after a major depressive episode - A 3-wave population-based study. Archives of General Psychiatry, 61(10), 990-996

Owens, M., Herbert, J., Jones, P. B., Sahakian, B. J., Wilkinson, P. O., Dunn, V. J., … Goodyer, I. M. (2014). Elevated morning cortisol is a stratified population-level biomarker for major depression in boys only with high depressive symptoms. *Proceedings of the National Academy of Sciences of the United States of America*, *111*(9), 3638–3643

Plamer, C.A., Alfano, C.A., (2017) Sleep and emotion regulation: an organizing, integrative review, *Sleep Medicine Review,* 31, 6-16

Panda, S., Hogenesch, J.B., and Kay, S.A., (2002) Circadian rhythms from flies to human, *Nature*, 417(6886), 329-35

Petersen, A.C., Crockett, L., Richards, M., and Boxer, A., (1987) A self-report measure of pubertal status: reliability, validity and initial norms, *Journal of Youth and Adolescence*, 17(2), 117-33

Picci, G., Scherf, K.S., 2016. From caregivers to peers: Puberty shapes human face perception. Psychol. Sci. 27 (11), 1461–1473.

Pilcher, J.J., Ginter D.R., and Sadowsky, B., (1997) Sleep quality versus sleep quantity: relationships between sleep and measures of health, well-being and sleepiness in college students, *Journal of Psychosomatic Research*, 42(6), 583-96

Platt, B., Waters, A.M., Schulte-Koerne, G., Engelmann, L., Salemink, E., 2017. A Review of Cognitive biases in youth depression: attention, interpretation and memory. *Cognit. Emot.* 31 (3), 462–483.

Potter, G. D., Skene, D. J., Arendt, J., Cade, J. E., Grant, P. J., and Hardie, L. J. (2016). Circadian rhythm and sleep disruption: causes, metabolic consequences, and countermeasures. *Endocrinology Review*.37, 584–608.

Power, R.A., and Pluess, M., (2015) Heritability estimates of the big five personality traits based on common genetic variants, *Translational Psychiatry*, 5(e604)

Power, M.J., Katz, R., McGuffin, P., Duggan, C.F., Lam, D., Beck, A.T., 1994. The Dysfunctional Attitude Scale (DAS): a comparison of forms A and B and proposals for a new subscaled version. *Journal of Research in Personality* 28 (3), 263–276.

Preussner, J.C., Kirschbaum, C., Meinlschmid, G., Hellhammer, D.H., (2003) Two formulas for computation of the area under the curve represent measures of total hormone concentration versus time dependent change, *Psychoneuroendocrinology*, 28(7), 916-31

Raja S.N., McGee, R., Stanton, W.R., (1992) Perceived attachments to parents and peers and psychological well-being in adolescence, *Journal of Youth and Adolescence*, 21(4), 471-85

Randler, C., Schredl, M., and Gorits, A.S., (2017) Chronotype, sleep behaviour and the big five personality factors, *Sage Open*, 1-9

Reinberg, A., and Ashkenazi, I., (2003) Concepts in human biological rhythms, *Dialogues in Clinical Neuroscience,* 5(4), 327-42
Reinecke, M.A., and Rodgers, G.M., (2001) Dysfunctional attitudes and attachment style among clinically depressed adults, *Psychiatry and Behavioural Sciences,* 29(2), 129-41
Renner, F., Pennix, B.W.J.H., Peeters, F., Cuijpers, P., and Huibers, M.J.H., (2013) Two-year stability and change of neuroticism and extraversion in treated and untreated persons with depression: Findings from the Netherlands Study of Depression and Anxiety (NESDA), *Journal of Affective Disorders*, 150, 2, (201),
Riberio, J.D., Pease, J.L., Guiterrez, P.M., Silva, C., Bernet, R.A., Rudd M.D., and Joiner, T.E. Jr., (2012) Sleep problems outperform depression and hopelessness as cross-sectional and longitudinal predictors of suicidal ideation and behaviour in young adults in the military, *Journal of Affective Disorders*, 136(3), 743-50
Rickard, N.S., Chin, T-C., and Vella-Brodrick, D.A. (2016) Cortisol awakening response as an index of mental health and well-being in adolescents, *Journal of Happiness Studies*, 17(6), 2555-2568
Roberts, R.E., and Duong, H.T., (2014) The prospective association between sleep deprivation and depression among adolescents, *Sleep*, 37(2), 239-244
Roberts, R.E., Lewinsohn, P.M., and Seeley, J.R., (1995) Symproms of DSM-III-R major depression in adolescence: evidence from an epidemiological survey, *Journal of the American Academy of Child and Adolescent Psychiatry*, 34(12), 1608-17
Robinson, M.S., Alloy, L.B., (2003). Negative cognitive styles and stress-reactive rumination interact to predict depression: a prospective study. *Cognitive Therapy Research* 27 (3), 275–291
Roenneberg, T., Kuehnle, T., Pramstaller, P.P., Ricken, J., Havel, M., Guth, A., and Merrow, M., (2004) A marker for the end of adolescence, *Current Biology*, 24(29),
Rogol, A.D., Roemmich, J.N., and Clark, P.A., (2002) Growth at puberty, *Journal of Adolescent Health*, 31(6), 192-200
Rosenthal, N.L., and Kobak, R., (2010) Assessing adolescents' attachment hierarchies: differences across developmental periods and associations with individual adaptation, *Journal of Research in Adolescence, 20(3), 678-706*
Rude, S.S., Wenzlaff, R.M., Gibbs, B., Vane, J., Whitney, T., 2002. Negative processing biases predict subsequent depressive symptoms. *Cognition and Emotion* 16 (3), 423–440.
Ryff, C. D., Singer, B. H., and Dienberg Love, G. (2004). Positive health: connecting well-being with biology. *Philosophical Transactions of the Royal Society B: Biological Sciences*, *359*(1449), 1383–1394.
Sadler, M.E., Miller, C.J., Christensen, K., McGue, M., 2011. Subjective well-being and longevity: a cotwin control study. *Twin Res. Hum. Genet. Off. J. Int. Soc. Twins* 14 (3), 249–256.
Sawyer, S.M., Afifi, R.A., Bearinger, L.H., Blakemore, S.J., Dick, B., Ezeh, A.C., Patton, G.C., (2012) Adolescence: a foundation for future health, *The Lancet*, 379(9826), 1630-40
Schepman, K., Taylor, E., Collishaw, S., Fombonne, E., 2011. Face emotion processing in depressed children and adolescents with and without comorbid conduct disorder. *Journal of Abnormal Child Psychology* 40 (4), 583–593.
Schmolke M (2003). Discovering positive health in the schizophrenic patient. A salutogenetic-oriented study. *Verhaltenstherapie* 13:102–109
Scottish Government (2017) Mental Health Strategy 2017-2027 – A 10-year vision, ISBN**:** 9781786526144, https://www.gov.scot/Publications/2017/03/1750/downloads#res-1
Shapero, B. G., Abramson, L. Y., & Alloy, L. B. (2016). Emotional Reactivity and Internalizing Symptoms: Moderating Role of Emotion Regulation. *Cognitive Therapy and Research*, *40*(30), 328–340. http://doi.org/10.1007/s10608-015-9722-4
Sharpley, C.F., (2009) Neurobiological pathways between chronic stress and depression: dysregulated adaptive mechanisms? *Clinical Medicine Insights: Psychiatry*, 2
Siegel, J.M., (2005) Clues to the functions of mammalian sleep, *Nature*, 437(7063), 1264-71
Smarr, B.L., and Schirmer, A.E., (2018) 3.4 million real-world learning management system logins reveal the majority of students experience social jet lag correlated with decreased performance, *Scientific Reports*, 8, 4793
Smith, E.M., Orchard, F., Reynolds, S., Whalley H.C., and Chan, S.W.Y., (2018), Cognitive biases predict symptoms of depression, anxiety and wellbeing above and beyond neuroticism in adolescence, *Journal of Affective Disorders,* 241:446-453,
Spence, S., 1998. A measure of anxiety symptoms among children. *Behaviour Research and Therapy* 3, 545–566.

Stalder, T., and Kirschbaum, C., (2012) Analysis of cortisol in hair – state of the art and future directions, *Brain, Behaviour and Immunity*, 26(7), 1019-29
Stalder, T., Kirschbaum, C., Kudielka, B.M., Adam, E.K., Pruessner, J.C., … Clow A., (2016) *Psychoneuroendocrinology,* 63, 414-32
Steinberg, L., (2005) Cognitive and affective development in adolescence *Trends in Cognitive Science*, 9(2), 69-74
Steinhausen H. C., Metzke C. W. (1998) Frequency and correlates of substance use among preadolescents and adolescents in a swiss epidemiological study. *J. Child Psych. Psych. Allied Dis,* 39: 387–397
Stephens, M.A., and Wand, G., (2012) Stress and the HPA axis: role of glucocorticoids in alcohol dependence, *Alcohol Research*, 34(4), 468-83
Steptoe, A., and Serwinski, B., (2016) Cortisol awakening response, In: Fink, G., (Eds.) *Stress: Concepts, Cognition, Emotion and Behaviour: Handbook of Stress*, 277-283
Studenmund, A.H., and Cassidy, H.J., (1987) *Using econometrics: a practical guide*. Boston: Little Brown
Suave, B., Koren, G., Walsh, G., Tokmakejian, S., and Van Uum, S.H., (2007) Measurement of cortisol in human hair as a biomarker of systemic exposure, *Clinical and Investigative Medicine*, 30(5), E183-91
Sumner, J.A., Griffith, J.W., Mineka, S., Rekart, K.N., Zinbarg, R.E., Craske, M.G., (2011) Overgeneral autobiographical memory and chronic interpersonal stress as predictors of the course of depression in adolescents. *Cognition and Emotion,* 25, 183–192.
Swales, M.A., Williams, J.M.G., Wood, P., (2010) Specificity of autobiographical memory and mood disorder in adolescents. *Cognition and Emotion,* 15 (3), 321–331.
Swendsen, J.D., and Merikangas, K.R., (2000) The comorbidity of depression and substance use disorders, *Clinical Psychology Review*, 20(2), 173-89
Talbot, L.S., McGlinchey, E.L., Kaplan, K.A., Dahl, R.E., and Harvey, A.G., (2010) Sleep deprivation in adolescents and adults: changes in affect, *Emotion*, 10(6), 831-41
Tang, T.Z., DeRubeis, R.J., Hollon, S.D., Amsterdam, J., Shelton, R., Schalet, B., (2009) A placebo-controlled test of the effects of paroxetine and cognitive therapy on personality risk factors in depression. *Archives of General Psychiatry* 66 (12), 1322–1330.
Tarokh, L., Saletin, J.M., and Carskadon, M.A., (2017) Sleep in adolescence: physiology, cognition and mental health, *Neuroscience and Biobehavioural Reviews*, 70, 182-188
Telzer, E. H., Goldenberg, D., Fuligni, A. J., Lieberman, M. D., & Gálvan, A. (2015). Sleep variability in adolescence is associated with altered brain development. *Developmental Cognitive Neuroscience, 14,* 16-22.
The Children and Young People (Scotland) Act *2014*, available at: http://www.legislation.gov.uk/asp/2014/8/contents/enacted
Thompson, B., (1995) Stepwise regression and stepwise discriminant analysis need not apply here: A guidelines editorial, *Educational and Psychological Measurement*, 55(4), 525-534
The National Sleep Foundation, (2000) *Adolescent Sleep Needs and Patterns*, Washington, D.C.
Timbremont, B., Braet, C., Bosmans, G., Van Vlierberghe, L., 2008. Cognitive biases in depressed and non-depressed referred youth. *Clinical Psychology and Psychotherapy* 15 (5), 329–339.
Traustadottir T, Bosch PR, Matt KS. Gender differences in cardiovascular and hypothalamic-pituitary-adrenal axis responses to psychological stress in healthy older adult men and women. *Stress*. 2003;6(2):133–140
Treynor, W., Gonzalez, R., Nolen-Hoeskema, S., 2003. Rumination Reconsidered: a psychometric analysis. *Cognit. Therapy Res*. 27 (3), 247–259.
Tsigos, C., Chrousos, G.P., (2002) Hypothalamic-pituitary-adrenal axis, neuroendocrine factors and stress, *Journal of Psychosomatic Research*, 53(4); 865-71
Tyron, W.W., (2004) Issues of validity in actigraphic sleep assessment, *Sleep*, 27(1) 158-65
Uher, R., and McGuffin, P., (2008) The moderation by the serotonin transporter gene of environmental adversity in the aetiology of mental illness: a review and methodological analysis, *Molecular Psychiatry*, 13(2), 131-46
Ulrike, S., Reinhold, L., and Dirk, H., (2013) Major depression in young girls is related to altered cortisol awakening response, *Child and Adolescent Psychiatry*, 22(6), 379-84
Urban, R., Magyarodi, T., and Rigo, A., (2011) Morningness-eveningness, chronotypes and health-impairing behaviors in adolescents, *Chronobiology International*, 28(3), 238-47
Urrila, A.S., Puanio, T., Palomaki, E., and Marttunen, M., (2015) Sleep in adolescent depression: physiological perspectives, *Acta Physiologica*, 213(4)
Van Uum, S.H., Suave, B., Fraser, L.A., Morley-Forster, P., Paul, T.L., and Koren, G., (2008) Elevated content of cortisol in hair of patients with severe chronic pain: a novel biomarker for stress, *Stress,* 11(6)483-8

Walker, E.R., McGee, R.E., and Druss, B.G., (2015) Mortality in mental disorders and global disease burden implications: a systematic review and meta-analysis, *JAMA Psychiatry*, 72(4), 334-41
Walters, E., and Cummings, E.M., (2000), A secure base from which to explore close relationships, *Child Development,* 71(1)
Wang M, Wright J, Brownlee A & Buswell R (2016) A comparison of approaches to stepwise regression on variables sensitivities in building simulation and analysis, *Computing Science and Mathematics Journal Articles,* 127, pp. 313-326.
Wang, P.S., Berglund, P., Olfson, M., Pincus, H.A., Wells, K.B., Kessler, R.C., 2005. Failure and delay in initial treatment contact after first onset of mental disorders in the new comorbidity survey replication. *Archives of General Psychiatry* 62 (2), 603–613.
Weiss, A. R., Johnson, N. L., Berger, N. A., and Redline, S. (2010). Validity of Activity-Based Devices to Estimate Sleep. *Journal of Clinical Sleep Medicine: JCSM: Official Publication of the American Academy of Sleep Medicine*, *6*(4), 336–342.
Weissman, M. M., Berry, O. O., Warner, V., Gameroff, M. J., Skipper, J., Talati, A., … Wickramaratne, P. (2016). A 30-Year Study of 3 Generations at High Risk and Low Risk for Depression. *JAMA Psychiatry*, *73*(9), 970–977
Whitmore, L.M., and Smith, T.C., (2018) Isolating the association of sleep, depressive state, and other independent indicators for suicide ideation in United States teenagers, *Archives of Suicide Research*, 23, 1-27
Wilhelm, I., Born, J., Kudielka, B.M., Scholts, W., and Wust, S., (2007) Is the cortisol awakening rise a response to awakening? *Psychoneuroendocrinology*, 32(4), 358-66
Williams, J.M.G., Broadbent, K., 1986. Autobiographical memory in suicide attempters. *Journal of Abnormal* Psychology, 95, 144–149.
Williams, J.M.G., Watts, F.N., MacLeod, C., Matthews, A., 1997. Cognitive Psychology and Emotional Disorders (second edition). Wiley and Sons, England.
Windle, M. (1994). Temperamental inhibition and activation: Hormonal and psychosocial correlates and associated psychiatry disorders. *Personality and Individual Differences,* 17, 61–70.
Wittmann, M., Dinisch, J., Merrow, M., and Roenneberg, T., (2006) Social Jetlag: misalignment of biological and social time, *Journal of Biological and Medical Rhythm Research*, 23(1-2)
World Health Organization. (1997). WHOQOL Measuring Quality of Life. Geneva: World Health Organisation.
World Health Organisation, (2013), Mental Health Action Plan: 2013-2020, ISBN: 978 92 4 150602.
World Health Organisation, 2017. Depression Fact Sheet.http://www.who.int/mediacentre/factsheets/fs369/en/
Wray, N.R., Ripke, S., … the Major Depressive Disorder Working Group of the Psychiatric Genomics Consortium, (2017) Genome-wide association analyses identify 44 risk ariants and refine the genetic architecture of major depression, *Nature Genetics*, 50, 668-681
Wray, N.R., Birley, A.J., Sullivan, P.F., Visscher, P.M., Martin, (2007). Genetic and phenotypic stability of measures of neuroticism over 22 years. *Twin Research and Human Genetics* 10 (5), 695–702.
Yamada, J., Stevens, B., de Silva, N., Gibbins, S., Beyene, J., Taddio, A., Newman, C., and Koren, G., (2007) Hair cortisol as a potential biologic marker of chronic stress in hospitalised neonates, *Neonatology,* 92(1), 42-9
Yoo, S.S., Gujar, N., Hu, P., Jolesz F.A., and Walker, M.P., (2007) The human emotional brain without sleep – a prefrontal amygdala disconnect, *Current Biology,* 17(20), R877-8
Young, C.C., Dietrich, M.S., 2015. Stressful life events, worry and rumination predict depressive and anxiety symptoms in young adolescents. *Journal of Adolescent Psychiatric Nursing*. 28 (1), 35–42.
Youth Access/ The Young People's Health Partnership (2017) Policy Briefing: March 2017, *Young, adult- and ignored: Getting a fair deal for 16-24 year olds from mental health services*, http://www.youngpeopleshealth.org.uk/wp-content/uploads/2017/03/Young-adult-and-ignored.pdf
Zamanian, Z., Dehghani, M., & Hashemi, H. (2013). Outline of Changes in Cortisol and Melatonin Circadian Rhythms in the Security Guards of Shiraz University of Medical Sciences. *International Journal of Preventive Medicine*, *4*(7), 825–830.
Zupan, B.A., Hammen, C., Jaenicke, C., 1987. The effects of current mood and prior depressive history on self-schematic processing in children. *Journal of Experimental Child Psychology*, 43 (1), 149–158.

Websites

RN Labs – DUTCH PLUS, cortisol awakening response testing
https://practitioner.researchnutrition.com.au/functional-tests/dutch-plus/

accessed 19/09/2018

Salimetrics Salivary Cortisol Enzyme Immunoassay Kit, (2016) www.Salimetrics.com

Scottish Index of Multiple Deprivation, https://www.gov.scot/Topics/Statistics/SIMD

Appendix

Chapter Two and Chapter Three Documents
- Ethical approval – Edinburgh University Ethics
- Ethical approval – Edinburgh City Council Ethics
- Information Sheets – Parental Information Sheet
- Information Sheet– Participant Information Sheet
- Consent Form – Parental Consent
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Chapter Four and Chapter Five Documents
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Chapter Five Results
- Gender Differences
- Full Regression Results

SCHOOL *of* HEALTH IN SOCIAL SCIENCE
CLINICAL PSYCHOLOGY

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Eilidh Smith
MScR Student
Clinical and Health Psychology
University of Edinburgh

25 February 2015

Dear Eilidh,

Application for Level 2/3 Approval

Re: Cognitive Functioning and Mood in Adolescents

Thank you for submitting the above research project for review by the Section of Clinical Psychology Ethics Research Panel. I can confirm that the submission has been independently reviewed and was approved on the 24th February 2015.

Should there be any change to the research protocol it is important that you alert us to this as this may necessitate further review.

Yours sincerely,

Kirsty Gardner

Administrator
Clinical Psychology

Eilidh Smith

Date	26 February 2015
Your ref	
Our ref	SCS/RW
Direct dial	0131 469 3137

Dear Eilidh,

I am writing in response to your application requesting permission to undertake research in The City of Edinburgh.

Your request has been considered, and I am pleased to inform you that you have been given permission in principle to undertake your research. I must stress that it is the policy of this Authority to leave the final decision about participation in research projects of this kind to Head Teachers and their staff, so that approval in principle does not oblige any particular establishment to take part.

I request that you forward a copy of your completed findings to me when they become available. In this case an electronic summary of your thesis would be preferred. Your work may be of interest to a number of staff in the Children and Families Department.

I would like to thank you for contacting the Children and Families Department about your work, and wish you every success in the completion of your project.

Yours sincerely

Ron Waddell
DSM and School Support Manager

Mood and Thinking
Information Sheet for Participants

Invitation to Participate in this Study
Thank you for your interest in taking part. Before deciding if you want to participate, it is important you understand what this research involves. Please take some time to read all the information below and feel free to ask any questions.

What are we trying to find out?
Every day we come across many different situations; there are different ways to interpret these situations. This study wants to see if how we think and feel about ourselves affects how we interpret situations. A second aspect of this study is memory; we want to see if mood has an effect on what we remember.

Who is organising the research?
This research is organised by Eilidh Smith, a postgraduate student at the Department of Clinical Psychology at the University of Edinburgh who is being supervised by Dr Stella Chan, a research fellow in the department.

Who can take part?
We are looking for young people aged 13-19 to take part.

Do I have to take part?
No, taking part is completely voluntary and you are free to change your mind and stop participating at any time without having to give any reason.

What will happen?
If you decide to take part, you will complete some questionnaires online or in person if you prefer. Then you will meet with Eilidh at an arranged time to complete some further tasks including memory and interpretation tasks. Completion of all tasks will take approximately one hour.

Are there any risks to taking part?
The tasks and questionnaires that you will be asked to complete have frequently been used in research so we do not think there are any risks to you taking part. However, you are free to stop participating at any point without having to give us a reason.

What are the benefits of taking part?
This study will increase understanding of how young people think and feel. You will also receive £10 of Amazon gift vouchers, as thank you for taking part.

Will my taking part in this research be kept a secret?
Yes, all the information we collect from you is confidential. You will not be asked to write your name on any of the questionnaires or tasks, instead you will be given a participant ID number. All the information collected will be kept safely for five years and then destroyed securely. The findings of this study will be written up as part of

Eilidh's thesis and may be published in academic journals or presented in conferences but your name will not be disclosed.

Audio Recording
As part of this research an audio recorder will be used to record the 'Autobiographical Memory Task'. This is to ensure that your response is detailed accurately. This is the only task that will involve the use of an audio recorder. After you have participated, the researcher will transcribe the audio recording and delete the audio file. Only the researcher and supervisor will have access to the audio recording prior to its deletion; to ensure this the audio recorder being used is fully encrypted.

Who has reviewed the research?
This research has been approved by the Research Ethics Committee of the School of Health in Social Science, University of Edinburgh.

What if there is a problem?
If you have any concerns please contact Eilidh Smith (see contact details below) and we will do our best to answer your questions. If you are still unhappy and would like to raise a formal complaint, please contact the Research Ethics Committee of the School of Health in Social Science, The University of Edinburgh (Tel: 0131 651 3969; Address: Medical Quad, Teviot Place, Edinburgh, EH8 9AG). You may also find it helpful to discuss this with parents, guardians or teachers.

How can I take part in this research or find out more about it?
If you decide to take part, we will ask you to sign a consent form to demonstrate your agreement to participate. If you are 15 years old or younger, we will also ask your parents or guardians to sign a consent form to show that they have agreed for you to participate and we will provide them with a separate Information Sheet so that they know what it is about. If you or your parents have any questions about the study at any point then please contact Eilidh Smith. Thank you for your interest in this study.

Eilidh Smith, Postgraduate research student in Clinical Psychology at the School of Health in Social Science, The University of Edinburgh, EH8 9AG
Email: *Eilidh.smith@ ed.ac.uk*

Dr Stella Chan, Chancellor's Fellow in Clinical Psychology,
School of Health in Social Science, The University of Edinburgh, EH8 9AG

Email: *stella.chan@ed.ac.uk*

Mood and Thinking

Information Sheet for Parents/Guardians

Dear Parent or Guardian,

Your child has expressed an interest in participating in our research study. As your child is under the age of 16, we must obtain parental consent before we can include your child in this study. Before deciding if you want your child to participate it is important that you understand what this research involves. Please take time to read the information below. *Please also remember that you and your child have no obligation to take part in this study.*

What are we trying to find out?
This study aims to find out whether the way we think and feel about ourselves affects how we interpret situations, facial expression and what we remember. We are particularly interested to see how this may affect emotional wellbeing of young people.

Who is organising the research?
This research is organised by Eilidh Smith, a postgraduate research student at the Department of Clinical Psychology at the University of Edinburgh, who is being supervised by Dr Stella Chan, who is an Academic Clinical Psychologist.

Who can take part?
We are recruiting young people between the age of 13 and 19.

What will happen if my child takes part?
Eilidh Smith will meet with your child at school, and they will be asked to complete some questionnaires about their mood and thinking styles, as well as some computer tasks that look at their memory and interpretation of emotional materials (e.g. facial expressions, everyday scenarios). Some tasks can be conducted online and if your child would prefer to complete these online this can be done prior to meeting Eilidh for the face-to-face tasks. The full set of tasks will take about an hour.

Are there any possible risks to my child?
All tasks and questionnaires have been frequently used in research and so we do not think there will be any problems or risks for your child. However, your child is free to withdraw from the study at any time without having to give any reason.

What are the possible benefits of taking part?

This study will help us better understand how young people think. As a small token of thanks, your child will receive £10 on completion of the study.

Will information be kept confidential?
Yes, all the information we collect from your child is confidential. We will not ask your child to write his/her name in any questionnaire or computer task, instead your child will be given a participant ID number. The consent form containing their name will be stored separately from the other information gathered. Information will be kept securely in locked offices in the University or password protected computers for up to five years after which it will be destroyed securely. The findings of the study will be written up as part of Eilidh Smith's research thesis and may be published in academic journals or presented at academic conferences. However, personal identifiable information of participants will not be disclosed.

Who has reviewed the research?
This research has been approved by the Research Ethics Committee of the School of Health in Social Science, University of Edinburgh.

What if there is a problem?
If you, or your child, are worried about anything to do with the research please contact Eilidh Smith or her supervisor Dr Stella Chan (see contact information below). We will do our best to answer your questions. If you are still unhappy and would like to raise a formal complaint, please contact the Research Ethics Committee of the School of Health in Social Science, The University of Edinburgh (Tel: 0131 651 3969; Address: Medical Quad, Teviot Place, Edinburgh, EH8 9AG).

How can my child take part in this research of find out more about it?
If you are happy for your child to participate in this research please give consent on the consent form provided. IF you or your child has any questions about the study, please contact Eilidh Smith. Thank you for your interest in this research project.

Eilidh Smith, Postgraduate research student of Clinical Psychology, School of Health in Social Science, The University of Edinburgh, EH8 9AG.
Email*: s1028516@sms.ed.ac.uk*

Dr Stella Chan, Chancellor's Fellow in Clinical Psychology, School of Health in Social Science, The University of Edinburgh, EH8 9AG
Email: *stella.chan@ed.ac.uk*

CONSENT FORM FOR PARTICIPANTS (aged 16+)

Title of Project: Mood and Thinking
Name of Investigator: Eilidh Smith (Researcher)
Stella Chan (Supervisor)

Please initial in the box

1. I have read and understood the information sheet for the above research and have had the opportunity to consider the information, ask questions and have received satisfactory answers. ☐
2. I agree to the use of an audio recorder for the 'Autobiographical Memory Task'. ☐
3. I understand that my participation is voluntary and that I am free to withdraw at any time without having to give a reason.
4. I confirm that I am 16 years old or above, and that I agree to take part in the above research ☐

Name of Participant	Signature	Date

Name of Researcher	Signature	Date

CONSENT FORM FOR PARENTS/GUARDIANS

Title of Project: Mood and Thinking
Name of Investigator: Eilidh Smith (Researcher)
Stella Chan (Supervisor)

Please initial in the box

1. I have read and understood the information sheet for the above research and have had the opportunity to consider the information, ask questions and have received satisfactory answers. ☐

2. I understand that my child's participation is voluntary and that she/he can withdraw at any time without having to give a reason. I also understand that I can withdraw my child from the study at any time. ☐

3. I agree for my child to take part in the above research ☐

Name of child:

Signature of Parent/Guardian:

Date:

ASSENT FORM FOR PARTICIPANTS (under 16)

Title of Project: Mood and Thinking
Name of Investigator: Eilidh Smith (Researcher)
Stella Chan (Supervisor)

Please initial in the box

1. I have read and understood the information sheet for the above research and have had the opportunity to consider the information, ask questions and have received satisfactory answers. ☐

2. I understand that my participation is voluntary and that I am free to withdraw at any time without having to give a reason. ☐

3. I agree to take part in the above research ☐

Name of Participant

Signature

Date

Name of Researcher

Signature

Date

FUTURE CONTACT

Thank you for participating in our study 'Mood and Thinking in Adolescence'.

We are interested in conducting further research to explore how mood and thinking may change over time. If you would be willing to be contacted again in the future, then please leave your contact information on this form.

By signing this form you will allow the researchers to contact you in the future to give you information about upcoming studies and to ask if you would like to participate. **However, this form does <u>not</u> obligate you to actually participate in any future studies.**

If you sign this form then the researcher will keep your contact details securely, unless you withdraw your permission by contacting the researchers.

If you agree to be contacted in the future, please indicate your preferred contact method and sign below.

Preferred contact method:

☐ phone:

_____

☐ post:

_____

☐ e-mail

Address:_____

_____ _____

Signature Date

_____ _____

Signature of Researcher Date

Mood and Thinking

Debrief Form

Thank you very much for participating!

This study is concerned with understanding the relationship between mood and styles of thinking, interpreting and remembering. Previous studies have found that people who experience distress demonstrate certain biases in memory and interpretation and are likely to experience unhelpful thinking styles that can further increase their distress.

Most of previous research has involved adults who have been diagnosed with mood disorders. As less research has involved young people from the general population, this study has targeted this group. Also, adolescence is an age where there is an increase of mood difficulties and so it is important to understand why this happens. We hope to increase the understanding of mood and thinking styles in young people, increasing those who experience distress and those who do not.

If you feel that you are having difficulties with your own mood, it is important that you talk to somebody about this. School guidance counsellors or your GP can advise you of how to access support. You may also find it helpful to talk to your parents.

If you have any questions about this research, please contact us:

Eilidh Smith, Postgraduate research student in Clinical Psychology at the School of Health in Social Science, The University of Edinburgh, EH8 9AG
Email: *s1028516@sms.ed.ac.uk*

Supervised by Dr Stella Chan, Chancellor's Fellow in Clinical Psychology, School of Health in Social Science, The University of Edinburgh, EH8 9AG
Email: *stella.chan@ed.ac.uk*

Thank you again for your participation.

·EDINBVRGH·
THE CITY OF EDINBURGH COUNCIL

CHILDREN'S SERVICES

Eilidh Smith
Postgraduate Researcher
School of Health in Social Science
University of Edinburgh
Doorway 6, Medical Quad
Teviot Place
Edinburgh
EH8 9AG

Date 03/11/16

Your ref

Our ref MG/AF

Dear Eilidh,

I am writing in response to your application requesting permission to undertake research in schools in The City of Edinburgh.

Your request has been considered, and I am pleased to inform you that you have been given permission in principle to undertake your research. I must stress that it is the policy of this Authority to leave the final decision about participation in research projects of this kind to Head Teachers and their staff, so that approval in principle does not oblige any particular establishment to take part.

I request that you forward a copy of your completed findings to me when they become available. In this case an electronic summary of your thesis would be preferred. Your work may be of interest to a number of staff in the Communities and Families Department.

I would like to thank you for contacting the Communities and Families Department about your work, and wish you every success in the completion of your project.

Yours sincerely

Martin Gemmell
Principal Psychologist

Psychological Services, Children's Services
Level 1.9 Waverley Court, 4 East Market Street, Edinburgh EH8 8BG

Tel 0131 469 2803 E-mail anne.fitzpatrick@ea.edin.sch.uk

INVESTORS IN PEOPLE | Gold

SCHOOL *of* HEALTH IN SOCIAL SCIENCE
CLINICAL AND HEALTH PSYCHOLOGY

The University of Edinburgh
Medical School
Doorway 6, Teviot Place
Edinburgh EH8 9AG

Telephone 0131 651 3969
Fax 0131 650 3891
Email submitting.ethics@ed.ac.uk

Eilidh Smith
PhD Student
Department of Clinical and Health Psychology
University of Edinburgh

31 August 2016

Dear Eilidh,

Application for Level 2 Approval

Reference: CLIN310
Project Title: The role of biological stress response and sleep in adolescents with or without low mood / Lay: How Well Do You Sleep?
Academic Supervisor: Stella Chan / Heather Whalley

Thank you for submitting the above research project for review by the Department of Clinical and Health Psychology Ethics Research Panel. I can confirm that the submission has been independently reviewed and was approved on the 13th August 2016.

Should there be any change to the research protocol it is important that you alert us to this as this may necessitate further review.

Yours sincerely,

Kirsty Gardner

Administrator
Clinical Psychology

Stress, Sleep and Mood
Information Sheet for Participants

Invitation to Participate in this Study
Thank you for your interest in taking part. Before deciding if you want to participate, it is important you understand what this research involves. Please take some time to read all the information below and feel free to ask any questions.

What are we trying to find out?
Mood and sleep can have an effect on our biology. Cortisol is considered to be a 'stress hormone' and can be affected by things like mood and sleep. Emotional regulation is the way in which we internally control the impact our emotions have on us. For example, distracting yourself by doing an activity if you are feeling anxious, or doing something you enjoy if you are feeling sad. This study hopes to investigate the relationships between these factors to try to better understand the impact or causes of different mood states.

Who is organising the research?
This research is organised by Eilidh Smith, a PhD student at the Department of Clinical Psychology at the University of Edinburgh who is being supervised by Dr Stella Chan, a research fellow in the department.

Who can take part?
We are looking for young people aged 13-19 to take part.

Do I have to take part?
No, taking part is completely voluntary and you are free to change your mind and stop participating at any time without having to give any reason.

What will happen?
If you decide to take part, you will meet with Eilidh who will explain the study in more detail. You will be asked to complete some questionnaires online (or in person if you prefer) and you will be given a diary to take home and record details of your sleep when you wake up in the morning. You will also be given a wristwatch to wear at night, which will record your movement to give us more information about how well you slept that night. This shouldn't interrupt your sleep.

Cortisol levels will be measured through a small sample of your hair (this will be cut from halfway down the back of your head so it wont be visible) and from two samples of saliva. Then you will meet with Eilidh again to return the sleep diary and saliva samples.

Are there any risks to taking part?
The questionnaires and measures involved have frequently been used in research so we do not think there are any risks to you taking part. However, you are free to stop participating at any point without having to give us a reason.

What are the benefits of taking part?
This study aims to increase understanding of how mood impacts young people. You will also receive £10 of Amazon gift vouchers, as thank you for taking part.

Will my taking part in this research be kept a secret?

Yes, all the information we collect from you is confidential. You will not be asked to write your name on any of the questionnaires or tasks, instead you will be given a participant ID number, this is so that all the information collected is anonymous. All the information collected will be kept safely for up to ten years and then destroyed securely. The findings of this study will be written up as part of Eilidh's thesis and may be published in academic journals or presented in conferences but your name will not be disclosed.

Who has reviewed the research?
This research has been approved by the Research Ethics Committee of the School of Health in Social Science, University of Edinburgh.

What if there is a problem?
If you have any concerns please contact the researcher or supervisor (see contact details below) and we will do our best to answer your questions. You may also find it helpful to discuss this with parents, guardians or teachers.
If you are still unhappy and would like to raise a formal complaint, please contact the Professor Charlotte Clarke, the Head of the School of Health in Social Science, The University of Edinburgh (Tel: 0131 651 3969; Address: Medical Quad, Teviot Place, Edinburgh, EH8 9AG email: hos.health@ed.ac.uk). Alternatively, complete this online complaint form:
http://www.ed.ac.uk/files/imports/fileManager/WEB%20Complaint%20Form.pdf .

How can I take part in this research or find out more about it?
If you decide to take part, we will ask you to sign a consent form to demonstrate your agreement to participate. If you are 15 years old or younger, we will also ask your parents or guardians to sign a consent form to show that they have agreed for you to participate and we will provide them with a separate Information Sheet so that they know what it is about. If you or your parents have any questions about the study at any point then please contact Eilidh Smith. Thank you for your interest in this study.

Eilidh Smith, PhD research student of Clinical Psychology at the School of Health in Social Science, The University of Edinburgh, EH8 9AG
Email: *eilidh.smith@ed.ac.uk*
Dr Stella Chan, Chancellor's Fellow in Clinical Psychology, School of Health in Social Science, The University of Edinburgh, EH8 9AG

Email: *stella.chan@ed.ac.uk*

How Well Do You Sleep?
Information Sheet for Parents

Dear Parent or Guardian,

Your child has expressed an interest in participating in our research study. As your child is under the age of 16, we must obtain parental consent before we can include your child in our study. Before deciding if you would like your child to participate it is important that you understand what this research involves. Please take time to read the information below. *Please also remember that you and your child have no obligation to take part.*

What are we trying to find out?
This study aims to examine the impact of wellbeing on sleep, stress and how young people regulate their emotions. We hope that this research will help us develop better ways to support young people in the future.

Who is organising the research?
This research is organised by Eilidh Smith, a PhD student at the Department of Clinical Psychology at the University of Edinburgh who is being supervised by Dr Stella Chan and Dr Heather Whalley.

Who can take part?
We are looking for young people aged 13-19 to take part.

What will happen if my child takes part?
Eilidh will meet with your child at school and explain the study in more detail. Your child will be asked to do the following:

1. Complete some questionnaires online (or in person if they/you prefer). This will take about 20 minutes.
2. Take home diary and record details of your child's sleep over one week. They will also be given a wristwatch to wear at night, which will record movement to give us more information about how well they sleep that night. This should not interrupt sleep or cause any discomfort.
3. To measure levels of cortisol (a stress hormone), we will collect a small sample of your child hair (this will be cut from halfway down the back of your head so it won't be visible). We will also ask your child to give two samples of saliva at home on the next morning.

Eilidh will meet with your child again in school to collect the diary and saliva samples.

Are there any risks to taking part?
The questionnaires and procedure involved have frequently been used in research so we do not think there are any risks to taking part. Collection of hair and saliva, and wearing a wristwatch, will not cause any discomfort. However, your child is free to withdraw at any time without having to give us a reason.

What are the benefits of taking part?

This study aims to increase understanding of how sleep and stress affect young people. Your child will also receive £10 of Amazon gift vouchers as thank you for taking part.

Will information be kept confidential?
Yes, all the information we collect from is confidential. Participants will not be asked to write their name on any of the questionnaires or tasks, instead they will be given a participant ID number. All the information collected will be kept safely for up to ten years and then destroyed securely. The findings of this study will be written up as part of Eilidh's thesis and may be published in academic journals or presented in conferences but names will not be disclosed.

Who has reviewed the research?
This research has been approved by the Research Ethics Committee of the School of Health in Social Science, University of Edinburgh.

What if there is a problem?
If you have any concerns please contact the researcher or supervisors (see contact details below) and we will do our best to answer your questions./ If you are still unhappy and would like to raise a formal complaint, please contact the Professor Charlotte Clarke, Head of the School of Health in Social Science, The University of Edinburgh (Tel: 0131 651 3969; Address: Medical Quad, Teviot Place, Edinburgh, EH8 9AG email: hos.health@ed.ac.uk). Alternatively, you can complete this online complaint form:
http://www.ed.ac.uk/files/imports/fileManager/WEB%20Complaint%20Form.pdf .

How can my child take part in this research or find out more about it?
If you are happy for your child to participate in this research, please give consent on the consent form provided. If you or your child have any questions about the study, please contact us (see contact details below). Thank you for your interest in this study.

Eilidh Smith, PhD research student of Clinical Psychology at the School of Health in Social Science, The University of Edinburgh, EH8 9AG

Email: *eilidh.smith@ed.ac.uk*

Supervised by Dr Stella Chan, Lecturer in Clinical Psychology, School of Health in Social Science, The University of Edinburgh, EH8 9AG

Email: *stella.chan@ed.ac.uk* Tel: 0131 651 3935

CONSENT FORM FOR PARTICIPANTS (aged 16+)

Title of Project: Stress, Sleep and Mood
Name of Investigator: Eilidh Smith (Researcher)

Stella Chan (Supervisor)

Please initial in the box

5. I have read and understood the information sheet for the above research and have had the opportunity to consider the information, ask questions and have received satisfactory answers. ☐

6. I understand that my participation is voluntary and that I am free to withdraw at any time without having to give a reason. ☐

7. I confirm that I am 16 years old or above, and that I agree to take part in the above research ☐

Name of Participant	Signature	Date

Name of Researcher	Signature	Date

CONSENT FORM FOR PARENTS/GUARDIANS

Title of Project: Stress, Sleep and Mood
Name of Investigator: Eilidh Smith (Researcher)
Stella Chan (Supervisor)

Please initial in the box

1. I have read and understood the information sheet for the above research and have had the opportunity to consider the information, ask questions and have received satisfactory answers. ☐
2. I understand that my child's participation is voluntary and that she/he can withdraw at any time without having to give a reason. I also understand that I can withdraw my child from the study at any time. ☐
3. I agree for my child to take part in the above research ☐

Name of child: _____

Signature of Parent/Guardian: _____

Date: _____

ASSENT FORM FOR PARTICIPANTS (under 16)

Title of Project: Stress, Sleep and Mood
Name of Investigator: Eilidh Smith (Researcher)
Stella Chan (Supervisor)

Please initial in the box

1. I have read and understood the information sheet for the above research and have had the opportunity to consider the information, ask questions and have received satisfactory answers. ☐
2. I understand that my participation is voluntary and that I am free to withdraw at any time without having to give a reason. ☐
3. I agree to take part in the above research ☐

Name of Participant	Signature	Date

Name of Researcher	Signature	Date

FUTURE CONTACT

Thank you for participating in our study ‘Stress, Sleep and Mood’.

We are interested in conducting further research to explore how mood impacts young people. If you would be willing to be contacted again in the future, then please leave your contact information on this form.

By signing this form you will allow the researchers to contact you in the future to give you information about upcoming studies and to ask if you would like to participate. **However, this form does <u>not</u> obligate you to actually participate in any future studies.**

If you sign this form then the researcher will keep your contact details securely, unless you withdraw your permission by contacting the researchers.

If you agree to be contacted in the future, please indicate your preferred contact method and sign below.

Preferred contact method:

Phone:_____

Post: _____

Email address:_____

_____ _____

Signature Date

_____ _____

Signature of Researcher Date

Stress, Sleep and Mood

Debrief Form

Thank you very much for participating!

This study is concerned with understanding the relationship between mood, sleep and levels of cortisol, a stress hormone. Previous studies have found that mood is often related to difference in sleep and biological measures of stress (cortisol).

Most of previous research has involved adults. As less research has involved young people from the general population, this study has targeted this group. Also, adolescence is an age where there is an increase of emotional difficulties and so it is important to understand why this happens. We hope to increase the understanding of how mood, cortisol levels and are associated in young people.

If you feel that you are having difficulties and would like to talk to somebody about this, you may find it helpful to speak to your parents, school guidance counsellors, or your GP.

YoungMinds is an organisation dedicated to providing education and support regarding young people's mental wellbeing. You may find their website of interest:

http://www.youngminds.org.uk/

If you have any questions or concerns about this research, please contact us:

Eilidh Smith, PhD research student in Clinical Psychology at the School of Health in Social Science, The University of Edinburgh, EH8 9AG
Email: *eilidh.smith@ed.ac.uk*

Supervised by Dr Stella Chan, Chancellor's Fellow in Clinical Psychology, School of Health in Social Science, The University of Edinburgh, EH8 9AG
Email: *stella.chan@ed.ac.uk*

School Academic Administrator, Emily Gribbin, School of Health in Social Science, The University of Edinburgh, EH8 9AG
Email: *emily.gribbin@ed.ac.uk*

Thank you again for your participation!

Chapter Five

Independent Samples Test										
MALE vs FEMALE	Levene's Test for Equality of Variances	t-test for Equality of Means								
		F	Sig.	t	df	Sig. (2-tailed)	Mean Difference	Std. Error Difference	95% Confidence Interval of the Difference	
									Lower	Upper
bed	Equal variances assumed	5.509	**0.021**	-0.839	91	0.404	-0:43	00:52	0	00:59
	Equal variances not assumed			-0.649	24.552	0.522	-0:43	01:07		01:35
sleep	Equal variances assumed	1.864	0.176	-1.364	91	0.176	#VALUE!	01:31		00:56
	Equal variances not assumed			-1.287	30.085	0.208	#VALUE!	01:36	0	01:13
sol	Equal variances assumed	0.989	0.323	-1.422	91	0.159	-5.40236	3.79969	-12.94998	2.14525
	Equal variances not assumed			-1.6	39.732	0.118	-5.40236	3.37638	-12.22772	1.42299
nawakenings	Equal variances assumed	1.091	0.299	-0.667	90	0.506	-0.13084	0.19604	-0.52031	0.25863
	Equal variances not assumed			-0.734	38.408	0.467	-0.13084	0.17814	-0.49133	0.22965
waso	Equal variances assumed	0.629	0.43	-0.273	90	0.785	-0.75603	2.76953	-6.25819	4.74612
	Equal variances not assumed			-0.352	54.213	0.726	-0.75603	2.14925	-5.06462	3.55256
wake	Equal variances assumed	0.015	0.903	0.174	89	0.862	00:02:36	00:14:58	-0:27:09	00:32:22
	Equal variances not assumed			0.195	36.568	0.847	00:02:36	00:13:23	-0:24:32	00:29:45
getup	Equal variances assumed	0.249	0.619	-0.693	91	0.49	-0:06:22	00:09:12	-0:24:39	00:11:53
	Equal variances not assumed			-0.653	30.042	0.519	-0:06:22	00:09:46	-0:26:19	00:13:34
MEANTST	Equal variances assumed	1.062	0.306	1.397	91	0.166	51.71627	37.02769	-21.83468	125.2672
	Equal variances not assumed			0.984	22.94	0.335	51.71627	52.56548	-57.03953	160.4721

Independent Samples Test						

	Levene's Test for Equality of Variances	t-test for Equality of Means								
		F	Sig.	t	df	Sig. (2-tailed)	Mean Difference			
								stedif	Lower	Upper
OnsetLatencyMean	Equal variances assumed	5.835	0.019	0.45	62	0.654	2.86448	6.36109	-9.85115	15.58012
Equal variances not assumed			0.333	19.835	0.743	2.86448	8.59869	-15.08162		20.81059
EfficiencyMean	Equal variances assumed	8.448	0.005	-0.18	62	0.858	-0.3403	1.88752	-4.11339	3.43279
Equal variances not assumed			-0.148	22.388	0.884	-0.3403	2.30137	-5.10826		4.42766
WASOMean	Equal variances assumed	0.908	0.344	-1.595	62	0.116	-8.43066	5.28464	-18.9945	2.13319
Equal variances not assumed			-1.733	37.308	0.091	-8.43066	4.86588	-18.28712		1.42581
PercentWakeMean	Equal variances assumed	0.004	0.952	-0.799	62	0.427	-0.76605	0.95908	-2.68323	1.15113
Equal variances not assumed			-0.745	27.261	0.463	-0.76605	1.02815	-2.8747		1.3426
SleepTimeMean	Equal variances assumed	0.215	0.645	-0.781	62	0.438	-21.94077	28.0774	-78.0667	34.18516
Equal variances not assumed			-0.758	29.255	0.455	-21.94077	28.96384	-81.15603		37.27449
PercentSleepMean	Equal variances assumed	0.004	0.952	0.799	62	0.427	0.76605	0.95908	-1.15113	2.68323
Equal variances not assumed			0.745	27.261	0.463	0.76605	1.02815	-1.3426		2.8747
actigraphawakeningsmean	Equal variances assumed	1.192	0.279	-1.569	62	0.122	-7.08313	4.51519	-16.10887	1.94261

Equal variances not assumed			-1.403	25.388	0.173	-7.08313	5.04962	-17.47496		3.3087
actbedtimenum	Equal variances assumed	0.061	0.806	0.736	62	0.464	0.011659	0.015834	-0.019993	0.04331
Equal variances not assumed			0.686	27.182	0.499	0.011659	0.017002	-0.023215		0.046532
AWAKETIME	Equal variances assumed	0.439	0.51	-1.236	62	0.221	-0.015747	0.012743	-0.04122	0.009726
Equal variances not assumed			-1.261	32.454	0.216	-0.015747	0.012486	-0.041167		0.009672

Hierarchical Regression Predicting MFQ with Sleep Diary Variables

Variables Entered/Removeda

Model	Variables Entered	Variables Removed	Method
1	PUBERTY, GENDER, AGEb	.	Enter
2	N.TOTALb	.	Enter
3	SDTIBNUM, LGSD5, SD2NUM, LGSD4, LGSD3, SD7NUM, SDTSTMINSb	.	Enter

a Dependent Variable: MFQ.TOTAL

b All requested variables entered.

Model Summary d										
Model	R	R Square	Adjusted R Square	Std. Error of the Estimate	Change Statistics					Durbin-Watson
					R Square Change	F Change	df1	df2	Sig. F Change	
1	.128 a	0.016	-0.017	14.4	0.016	0.492	3	88	0.689	
2	.612 b	0.375	0.346	11.55	0.358	49.801	1	87	0	
3	.741c	0.549	0.487	10.229	0.174	4.418	7	80	0	2.229
a Predictors: (Constant), PUBERTY, GENDER, AGE										
b Predictors: (Constant), PUBERTY, GENDER, AGE, N.TOTAL										

c Predictors: (Constant), PUBERTY, GENDER, AGE, N.TOTAL, SDTIBNUM, LGSD5, SD2NUM, LGSD4, LGSD3, SD7NUM, SDTSTMINS						
d Dependent Variable: MFQ.TOTAL						
ANOVAa						
Model		Sum of Squares	df	Mean Square	F	Sig.
1	Regression	306.018	3	102.006	0.492	.689b
	Residual	18248.896	88	207.374		
	Total	18554.913	91			
2	Regression	6949.35	4	1737.337	13.024	.000c
	Residual	11605.563	87	133.397		
	Total	18554.913	91			
3	Regression	10184.818	11	925.893	8.85	.000d
	Residual	8370.095	80	104.626		
	Total	18554.913	91			
a Dependent Variable: MFQ.TOTAL						
b Predictors: (Constant), PUBERTY, GENDER, AGE						
c Predictors: (Constant), PUBERTY, GENDER, AGE, N.TOTAL						
d Predictors: (Constant), PUBERTY, GENDER, AGE, N.TOTAL, SDTIBNUM, LGSD5, SD2NUM, LGSD4, LGSD3, SD7NUM, SDTSTMINS						

Coefficienta													
Model		Unstandardized Coefficients		Standard	t	Sig.	95.0% Confidence Interval for B		Correlations				
		B	Std. Error	Beta			Lower Bound	Upper Bound	Zero-order	Partial	Part	Tolerance	VIF
1	(Constant)	22.797	14.748		1.546	0.126	-6.511	52.105					
	AGE	-0.632	1.05	-0.075	-0.602	0.549	-2.719	1.456	-0.073	-0.064	-0.064	0.719	1.391
	GENDER	3.274	3.914	0.095	0.836	0.405	-4.505	11.053	0.111	0.089	0.088	0.865	1.157
	PUBERTY	0.144	0.645	0.029	0.224	0.824	-1.138	1.426	0.017	0.024	0.024	0.682	1.467

2	(Constant)	2.057	12.188		0.169	0.866	-22.168	26.282					
	AGE	0.168	0.85	0.02	0.198	0.844	-1.522	1.858	-0.073	0.021	0.017	0.706	1.416
	GENDER	1.807	3.146	0.052	0.574	0.567	-4.447	8.061	0.111	0.061	0.049	0.861	1.162
	PUBERTY	-0.283	0.521	-0.056	-0.544	0.588	-1.319	0.752	0.017	-0.058	-0.046	0.673	1.487
	N.TOTAL	2.418	0.343	0.609	7.057	0	1.737	3.099	0.609	0.603	0.598	0.964	1.037
3	(Constant)	-127.038	32.909		-3.86	0	-192.53	-61.546					
	AGE	-0.227	0.798	-0.027	-0.285	0.777	-1.815	1.361	-0.073	-0.032	-0.021	0.629	1.591
	GENDER	-0.537	2.966	-0.016	-0.181	0.857	-6.439	5.365	0.111	-0.02	-0.014	0.76	1.316
	PUBERTY	0.042	0.507	0.008	0.083	0.934	-0.967	1.051	0.017	0.009	0.006	0.557	1.796
	N.TOTAL	**2.224**	**0.316**	**0.561**	**7.031**	**0**	**1.595**	**2.854**	**0.609**	**0.618**	**0.528**	**0.887**	**1.127**
	SD2NUM	**93.314**	**34.378**	**0.221**	**2.714**	**0.008**	**24.899**	**161.728**	**0.3**	**0.29**	**0.204**	**0.85**	**1.176**
	LGSD3	**12.614**	**4.111**	**0.274**	**3.068**	**0.003**	**4.432**	**20.795**	**0.268**	**0.324**	**0.23**	**0.705**	**1.418**
	LGSD4	-5.363	3.602	-0.124	-1.489	0.14	-12.532	1.805	-0.165	-0.164	-0.112	0.81	1.234
	LGSD5	-0.801	2.192	-0.032	-0.365	0.716	-5.162	3.561	0.071	-0.041	-0.027	0.754	1.327
	SD7NUM	110.001	59.145	0.199	1.86	0.067	-7.701	227.702	0.277	0.204	0.14	0.49	2.039
	SDTIBNUM	4.158	10.858	0.031	0.383	0.703	-17.45	25.767	0.048	0.043	0.029	0.874	1.144
	SDTSTMINS	-0.021	0.025	-0.096	-0.851	0.397	-0.07	0.028	0.082	-0.095	-0.064	0.447	2.236

Hierarchical Regression Predicting SCAS with Sleep Diary Variables

Variables Entered/Removeda

Model	Variables Entered	Variables Removed	Method

1	PUBERTY, GENDER, AGEb	.	Enter
2	N.TOTALb	.	Enter
3	SDTIBNUM, LGSD5, SD2NUM, LGSD4, LGSD3, SD7NUM, SDTSTMINSb	.	Enter

a Dependent Variable: SCAS.TOTAL

b All requested variables entered.

Model Summary d										
Model	R	R Square	Adjusted R Square	Std. Error of the Estimate	Change Statistics					Durbin-Watson
					R Square Change	F Change	df1	df2	Sig. F Change	
1	.275a	0.076	0.044	19.817	0.076	2.405	3	88	0.073	
2	.745b	0.555	0.535	13.826	0.479	93.776	1	87	0	
3	.771c	0.594	0.539	13.769	0.039	1.104	7	80	0.369	2.37
a Predictors: (Constant), PUBERTY, GENDER, AGE										
b Predictors: (Constant), PUBERTY, GENDER, AGE, N.TOTAL										
c Predictors: (Constant), PUBERTY, GENDER, AGE, N.TOTAL, SDTIBNUM, LGSD5, SD2NUM, LGSD4, LGSD3, SD7NUM, SDTSTMINS										
d Dependent Variable: SCAS.TOTAL										

ANOVAa						
Model		Sum of Squares	df	Mean Square	F	Sig.
1	Regression	2833.618	3	944.539	2.405	.073b
	Residual	34558.067	88	392.705		
	Total	37391.685	91			
2	Regression	20760.278	4	5190.069	27.15	.000c
	Residual	16631.407	87	191.166		
	Total	37391.685	91			
3	Regression	22224.894	11	2020.445	10.657	.000d
	Residual	15166.791	80	189.585		
	Total	37391.685	91			
a Dependent Variable: SCAS.TOTAL						
b Predictors: (Constant), PUBERTY, GENDER, AGE						

c Predictors: (Constant), PUBERTY, GENDER, AGE, N.TOTAL				
d Predictors: (Constant), PUBERTY, GENDER, AGE, N.TOTAL, SDTIBNUM, LGSD5, SD2NUM, LGSD4, LGSD3, SD7NUM, SDTSTMINS				

Model		Unstandardized Coefficients		Standardized Coefficients	t	Sig.	95.0% Confidence Interval for B		Correlations			Collinearity Statistics	
		B	Std. Error	Beta			Lower Bound	Upper Bound	Zero-order	Partial	Part	Tolerance	VIF
1	(Constant)	35.289	20.294		1.739	0.086	-5.042	75.62					
	AGE	-1.863	1.446	-0.156	-1.289	0.201	-4.735	1.01	-0.101	-0.136	-0.132	0.719	1.391
	GENDER	8.435	5.387	0.173	1.566	0.121	-2.27	19.14	0.231	0.165	0.16	0.865	1.157
	PUBERTY	1.128	0.888	0.158	1.271	0.207	-0.636	2.892	0.128	0.134	0.13	0.682	1.467
2	(Constant)	1.219	14.59		0.084	0.934	-27.78	30.219					
	AGE	-0.549	1.018	-0.046	-0.539	0.591	-2.571	1.474	-0.101	-0.058	-0.039	0.706	1.416
	GENDER	6.025	3.767	0.123	1.6	0.113	-1.461	13.512	0.231	0.169	0.114	0.861	1.162
	PUBERTY	0.426	0.624	0.06	0.683	0.497	-0.814	1.665	0.128	0.073	0.049	0.673	1.487
	N.TOTAL	3.971	0.41	0.705	9.684	0	3.156	4.786	0.73	0.72	0.692	0.964	1.037
3	(Constant)	-75.986	44.3		-1.715	0.09	-164.146	12.173					
	AGE	-0.694	1.074	-0.058	-0.646	0.52	-2.831	1.444	-0.101	-0.072	-0.046	0.629	1.591
	GENDER	5.798	3.992	0.119	1.452	0.15	-2.147	13.742	0.231	0.16	0.103	0.76	1.316
	PUBERTY	0.49	0.682	0.069	0.718	0.475	-0.868	1.848	0.128	0.08	0.051	0.557	1.796
	N.TOTAL	**3.788**	**0.426**	**0.673**	**8.896**	**0**	**2.941**	**4.636**	**0.73**	**0.705**	**0.633**	**0.887**	**1.127**
	SD2NUM	58.574	46.277	0.098	1.266	0.209	-33.519	150.667	0.194	0.14	0.09	0.85	1.176
	LGSD3	1.634	5.534	0.025	0.295	0.769	-9.379	12.647	0.05	0.033	0.021	0.705	1.418
	LGSD4	-8.028	4.849	-0.131	-1.656	0.102	-17.678	1.622	-0.2	-0.182	-0.118	0.81	1.234
	LGSD5	-0.612	2.95	-0.017	-0.207	0.836	-6.483	5.26	0.032	-0.023	-0.015	0.754	1.327
	SD7NUM	112.552	79.616	0.144	1.414	0.161	-45.888	270.992	0.179	0.156	0.101	0.49	2.039
	SDTIBNUM	-5.313	14.616	-0.028	-0.363	0.717	-34.4	23.775	-0.021	-0.041	-0.026	0.874	1.144
	SDTSTMINS	-0.032	0.033	-0.102	-0.958	0.341	-0.098	0.034	0.103	-0.106	-0.068	0.447	2.236
a Dependent Variable: SCAS.TOTAL													

Hierarchical Regression Predicting BBC with Sleep Diary Variables

Variables Entered/Removeda

Model	Variables Entered	Variables Removed	Method
1	PUBERTY, GENDER, AGEb	.	Enter
2	N.TOTALb	.	Enter
3	SDTIBNUM, LGSD5, SD2NUM, LGSD4, LGSD3, MedSBJRating, SDTSTMINSb	.	Enter

a Dependent Variable: BBC.TOTAL

b All requested variables entered.

Model Summaryd										
Model	R	R Square	Adjusted R Square	Std. Error of the Estimate	Change Statistics					Durbin-Watson
					R Square Change	F Change	df1	df2	Sig. F Change	
1	.243a	0.059	0.027	12.434	0.059	1.838	3	88	0.146	
2	.606b	0.367	0.338	10.254	0.308	42.388	1	87	0	
3	.683c	0.466	0.393	9.819	0.099	2.126	7	80	0.05	2.114
a Predictors: (Constant), PUBERTY, GENDER, AGE										
b Predictors: (Constant), PUBERTY, GENDER, AGE, N.TOTAL										
c Predictors: (Constant), PUBERTY, GENDER, AGE, N.TOTAL, SDTIBNUM, LGSD5, SD2NUM, LGSD4, LGSD3, MedSBJRating, SDTSTMINS										
d Dependent Variable: BBC.TOTAL										

ANOVAa						
Model		Sum of Squares	df	Mean Square	F	Sig.
1	Regression	852.494	3	284.165	1.838	.146b
	Residual	13604.495	88	154.597		
	Total	14456.989	91			

2	Regression	5309.385	4	1327.346	12.624	.000c
	Residual	9147.604	87	105.145		
	Total	14456.989	91			
3	Regression	6744.077	11	613.098	6.359	.000d
	Residual	7712.912	80	96.411		
	Total	14456.989	91			
a Dependent Variable: BBC.TOTAL						
b Predictors: (Constant), PUBERTY, GENDER, AGE						
c Predictors: (Constant), PUBERTY, GENDER, AGE, N.TOTAL						
d Predictors: (Constant), PUBERTY, GENDER, AGE, N.TOTAL, SDTIBNUM, LGSD5, SD2NUM, LGSD4, LGSD3, MedSBJRating, SDTSTMINS						

		Unstandardized Coefficients		Standardized Coefficients	t	Sig.	95.0% Confidence Interval for B		Correlations			Collinearity Statistics	
		B	Std. Error	Beta			Lower Bound	Upper Bound	Zero-order	Partial	Part	Tolerance	VIF
1	(Constant)	88.46	12.733		6.947	0	63.155	113.765					
	AGE	-0.529	0.907	-0.071	-0.583	0.561	-2.332	1.273	-0.175	-0.062	-0.06	0.719	1.391
	GENDER	1.914	3.38	0.063	0.566	0.573	-4.803	8.63	0.019	0.06	0.059	0.865	1.157
	PUBERTY	-0.908	0.557	-0.204	-1.631	0.106	-2.015	0.198	-0.222	-0.171	-0.169	0.682	1.467
2	(Constant)	105.448	10.82		9.745	0	83.941	126.955					
	AGE	-1.184	0.755	-0.159	-1.569	0.12	-2.684	0.316	-0.175	-0.166	-0.134	0.706	1.416
	GENDER	3.115	2.793	0.102	1.115	0.268	-2.437	8.667	0.019	0.119	0.095	0.861	1.162
	PUBERTY	-0.558	0.462	-0.126	-1.207	0.231	-1.477	0.361	-0.222	-0.128	-0.103	0.673	1.487
	N.TOTAL	-1.98	0.304	-0.565	-6.511	0	-2.585	-1.376	-0.548	-0.572	-0.555	0.964	1.037
3	(Constant)	103.852	34.864		2.979	0.004	34.47	173.234					
	AGE	-0.949	0.755	-0.128	-1.258	0.212	-2.451	0.553	-0.175	-0.139	-0.103	0.648	1.544
	GENDER	4.28	2.837	0.141	1.509	0.135	-1.365	9.925	0.019	0.166	0.123	0.765	1.306
	PUBERTY	-0.522	0.484	-0.117	-1.079	0.284	-1.486	0.441	-0.222	-0.12	-0.088	0.563	1.776
	N.TOTAL	**-1.863**	**0.306**	**-0.532**	**-6.094**	**0**	**-2.471**	**-1.255**	**-0.548**	**-0.563**	**-0.498**	**0.875**	**1.142**
	SD2NUM	12.979	33.485	0.035	0.388	0.699	-53.659	79.616	-0.163	0.043	0.032	0.826	1.211
	LGSD3	**-9.017**	**3.863**	**-0.222**	**-2.334**	**0.022**	**-16.704**	**-1.329**	**-0.084**	**-0.253**	**-0.191**	**0.736**	**1.359**
	LGSD4	1.81	3.445	0.047	0.525	0.601	-5.046	8.666	0.12	0.059	0.043	0.816	1.225

	LGSD5	2.835	2.169	0.127	1.307	0.195	-1.482	7.152	0.019	0.145	0.107	0.709	1.41
	SDTIBNUM	-14.869	10.361	-0.125	-1.435	0.155	-35.488	5.749	-0.082	-0.158	-0.117	0.885	1.13
	SDTSTMINS	-0.033	0.019	-0.17	-1.7	0.093	-0.072	0.006	-0.287	-0.187	-0.139	0.667	1.499
	MedSBJRating	2.906	1.62	0.171	1.793	0.077	-0.319	6.131	0.258	0.197	0.146	0.735	1.36
a Dependent Variable: BBC.TOTAL													

Hierarchical Regression Predicting MFQ with ACTIGRAPH Variables

Variables Entered/Removeda

Model	Variables Entered	Variables Removed	Method
1	PUBERTY, GENDER, AGEb	.	Enter
2	N.TOTALb	.	Enter
3	ATSTNUM, WASOMean, OnsetLatencyMean, meanwaketimeacti, meanBedtimeacti, actigraphawakeningsmean, EfficiencyMean, PercentSleepMean, ATIBNUMb	.	Enter

a Dependent Variable: MFQ.TOTAL

b All requested variables entered.

Model Summaryd										
Model	R	R Square	Adjusted R Square	Std. Error of the Estimate	Change Statistics					Durbin-Watson
					R Square Change	F Change	df1	df2	Sig. F Change	
1	.170a	0.029	-0.021	13.417	0.029	0.583	3	59	0.628	
2	.619b	0.384	0.341	10.78	0.355	33.408	1	58	0	
3	.661c	0.437	0.288	11.208	0.053	0.517	9	49	0.855	2.167
a Predictors: (Constant), PUBERTY, GENDER, AGE										
b Predictors: (Constant), PUBERTY, GENDER, AGE, N.TOTAL										
c Predictors: (Constant), PUBERTY, GENDER, AGE, N.TOTAL, ATSTNUM, WASOMean, OnsetLatencyMean, meanwaketimeacti, meanBedtimeacti, actigraphawakeningsmean, EfficiencyMean, PercentSleepMean, ATIBNUM										
d Dependent Variable: MFQ.TOTAL										

ANOVAa

Model			Sum of Squares	df	Mean Square	F	Sig.
	1	Regression	314.944	3	104.981	0.583	.628b
		Residual	10621.659	59	180.028		
		Total	10936.603	62			
	2	Regression	4196.943	4	1049.236	9.029	.000c
		Residual	6739.66	58	116.201		
		Total	10936.603	62			
	3	Regression	4781.522	13	367.809	2.928	.003d
		Residual	6155.081	49	125.614		
		Total	10936.603	62			

a Dependent Variable: MFQ.TOTAL

b Predictors: (Constant), PUBERTY, GENDER, AGE

c Predictors: (Constant), PUBERTY, GENDER, AGE, N.TOTAL

d Predictors: (Constant), PUBERTY, GENDER, AGE, N.TOTAL, ATSTNUM, WASOMean, OnsetLatencyMean, meanwaketimeacti, meanBedtimeacti, actigraphawakeningsmean, EfficiencyMean, PercentSleepMean, ATIBNUM

Model		Unstandardized Coefficients		Standardized Coefficients	t	Sig.	95.0% Confidence Interval for B		Correlations			Collinearity Statistics	
		B	Std. Error	Beta			Lower Bound	Upper Bound	Zero-order	Partial	Part	Tolerance	VIF
1	(Constant)	32.156	17.396		1.848	0.07	-2.653	66.965					
	AGE	-0.506	1.251	-0.061	-0.404	0.687	-3.009	1.997	-0.116	-0.053	-0.052	0.724	1.382
	GENDER	3.744	4.225	0.126	0.886	0.379	-4.71	12.197	0.077	0.115	0.114	0.813	1.231
	PUBERTY	-0.62	0.848	-0.12	-0.732	0.467	-2.316	1.076	-0.101	-0.095	-0.094	0.613	1.632
2	(Constant)	14.484	14.306		1.012	0.316	-14.154	43.121					
	AGE	0.05	1.01	0.006	0.049	0.961	-1.971	2.07	-0.116	0.006	0.005	0.717	1.395
	GENDER	3.494	3.394	0.118	1.029	0.308	-3.3	10.289	0.077	0.134	0.106	0.813	1.231
	PUBERTY	-0.907	0.683	-0.175	-1.328	0.189	-2.273	0.46	-0.101	-0.172	-0.137	0.609	1.641
	N.TOTAL	2.178	0.377	0.599	5.78	0	1.424	2.933	0.597	0.605	0.596	0.988	1.012
3	(Constant)	-13.074	128.323		-0.102	0.919	-270.949	244.801					
	AGE	-0.228	1.372	-0.027	-0.166	0.869	-2.985	2.53	-0.116	-0.024	-0.018	0.42	2.383
	GENDER	4.863	3.889	0.164	1.25	0.217	-2.953	12.68	0.077	0.176	0.134	0.669	1.495
	PUBERTY	-1.006	0.831	-0.195	-1.211	0.232	-2.676	0.664	-0.101	-0.17	-0.13	0.445	2.248

	N.TOTAL	**2.193**	**0.414**	**0.603**	**5.301**	**0**	**1.362**	**3.024**	**0.597**	**0.604**	**0.568**	**0.887**	**1.128**
	meanBedtimeacti	5.00E-05	0	0.127	0.882	0.382	0	0	-0.034	0.125	0.095	0.556	1.798
	meanwaketimeacti	0	0	0.137	0.916	0.364	0	0.001	0.081	0.13	0.098	0.511	1.956
	OnsetLatencyMean	0.083	0.269	0.141	0.307	0.76	-0.457	0.623	-0.033	0.044	0.033	0.054	18.389
	EfficiencyMean	0.242	1.803	0.122	0.134	0.894	-3.381	3.865	0.128	0.019	0.014	0.014	72.302
	WASOMean	-0.059	0.255	-0.086	-0.23	0.819	-0.572	0.455	-0.188	-0.033	-0.025	0.083	12.02
	PercentSleepMean	-0.093	2.427	-0.024	-0.038	0.97	-4.969	4.783	0.203	-0.005	-0.004	0.029	34.831
	actigraphawakeningsmean	-0.054	0.165	-0.067	-0.329	0.744	-0.385	0.277	-0.18	-0.047	-0.035	0.276	3.626
	ATIBNUM	0.236	0.322	1.574	0.735	0.466	-0.41	0.882	0.067	0.104	0.079	0.003	399.115
	ATSTNUM	-0.221	0.326	-1.597	-0.679	0.5	-0.875	0.433	0.066	-0.097	-0.073	0.002	481.516
a Dependent Variable: MFQ.TOTAL													

Hierarchical Regression Predicting SCAS with ACTIGRAPH Variables

Variables Entered/Removeda

Model	Variables Entered	Variables Removed	Method
1	PUBERTY, GENDER, AGEb	.	Enter
2	N.TOTALb	.	Enter
3	ATSTNUM, WASOMean, OnsetLatencyMean, meanwaketimeacti, meanBedtimeacti, actigraphawakeningsmean, EfficiencyMean, PercentSleepMean, ATIBNUMb	.	Enter

a Dependent Variable: SCAS.TOTAL

b All requested variables entered.

Model Summaryd										
Model	R	R Square	Adjusted R Square	Std. Error of the Estimate	Change Statistics					Durbin-Watson
					R Square Change	F Change	df1	df2	Sig. F Change	
1	.233a	0.054	0.006	19.356	0.054	1.128	3	59	0.345	
2	.727b	0.528	0.496	13.79	0.474	58.244	1	58	0	
3	.782c	0.612	0.508	13.613	0.083	1.169	9	49	0.336	1.931

a Predictors: (Constant), PUBERTY, GENDER, AGE
b Predictors: (Constant), PUBERTY, GENDER, AGE, N.TOTAL
c Predictors: (Constant), PUBERTY, GENDER, AGE, N.TOTAL, ATSTNUM, WASOMean, OnsetLatencyMean, meanwaketimeacti, meanBedtimeacti, actigraphawakeningsmean, EfficiencyMean, PercentSleepMean, ATIBNUM
d Dependent Variable: SCAS.TOTAL

ANOVAa

Model		Sum of Squares	df	Mean Square	F	Sig.
1	Regression	1268.385	3	422.795	1.128	.345b
	Residual	22105.044	59	374.662		
	Total	23373.429	62			
2	Regression	12344.098	4	3086.025	16.228	.000c
	Residual	11029.331	58	190.161		
	Total	23373.429	62			
3	Regression	14293.183	13	1099.476	5.933	.000d
	Residual	9080.245	49	185.311		
	Total	23373.429	62			

a Dependent Variable: SCAS.TOTAL

b Predictors: (Constant), PUBERTY, GENDER, AGE

c Predictors: (Constant), PUBERTY, GENDER, AGE, N.TOTAL

d Predictors: (Constant), PUBERTY, GENDER, AGE, N.TOTAL, ATSTNUM, WASOMean, OnsetLatencyMean, meanwaketimeacti, meanBedtimeacti, actigraphawakeningsmean, EfficiencyMean, PercentSleepMean, ATIBNUM

Model		Unstandardized Coefficients		Standardized Coefficients	t	Sig.	95.0% Confidence Interval for B		Correlations			Collinearity Statistics	
		B	Std. Error	Beta			Lower Bound	Upper Bound	Zero-order	Partial	Part	Tolerance	VIF
1	(Constant)	23.799	25.095		0.948	0.347	-26.417	74.014					
	AGE	-1.238	1.805	-0.102	-0.686	0.495	-4.849	2.373	-0.016	-0.089	-0.087	0.724	1.382
	GENDER	5.81	6.095	0.134	0.953	0.344	-6.385	18.005	0.194	0.123	0.121	0.813	1.231
	PUBERTY	1.227	1.223	0.162	1.004	0.32	-1.219	3.674	0.164	0.13	0.127	0.613	1.632

2	(Constant)	-6.052	18.302		-0.331	0.742	-42.686	30.582					
	AGE	-0.3	1.292	-0.025	-0.232	0.817	-2.885	2.286	-0.016	-0.03	-0.021	0.717	1.395
	GENDER	5.389	4.342	0.124	1.241	0.22	-3.303	14.081	0.194	0.161	0.112	0.813	1.231
	PUBERTY	0.743	0.873	0.098	0.851	0.398	-1.005	2.492	0.164	0.111	0.077	0.609	1.641
	N.TOTAL	3.679	0.482	0.692	7.632	0	2.714	4.644	0.704	0.708	0.688	0.988	1.012
3	(Constant)	-84.269	155.861		-0.541	0.591	-397.483	228.945					
	AGE	-1.167	1.667	-0.096	-0.7	0.487	-4.516	2.182	-0.016	-0.1	-0.062	0.42	2.383
	GENDER	5.006	4.724	0.115	1.06	0.294	-4.487	14.499	0.194	0.15	0.094	0.669	1.495
	PUBERTY	0.207	1.009	0.027	0.205	0.839	-1.822	2.235	0.164	0.029	0.018	0.445	2.248
	N.TOTAL	**3.846**	**0.502**	**0.724**	**7.656**	**0**	**2.837**	**4.856**	**0.704**	**0.738**	**0.682**	**0.887**	**1.128**
	meanBedtimeacti	**0**	**0**	**0.257**	**2.155**	**0.036**	**0**	**0**	**0.087**	**0.294**	**0.192**	**0.556**	**1.798**
	meanwaketimeacti	0	0	0.055	0.444	0.659	-0.001	0.001	0.011	0.063	0.04	0.511	1.956
	OnsetLatencyMean	0.191	0.326	0.223	0.585	0.561	-0.465	0.847	-0.096	0.083	0.052	0.054	18.389
	EfficiencyMean	1.757	2.19	0.608	0.803	0.426	-2.643	6.157	0.187	0.114	0.071	0.014	72.302
	WASOMean	-0.084	0.31	-0.083	-0.269	0.789	-0.707	0.54	-0.127	-0.038	-0.024	0.083	12.02
	PercentSleepMean	-0.814	2.947	-0.145	-0.276	0.784	-6.736	5.109	0.187	-0.039	-0.025	0.029	34.831
	actigraphawakeningsmean	**0.423**	**0.2**	**0.358**	**2.114**	**0.04**	**0.021**	**0.825**	**-0.019**	**0.289**	**0.188**	**0.276**	**3.626**

	ATIBNUM	0.218	0.391	0.992	0.558	0.58	-0.567	1.003	-0.015	0.079	0.05	0.003	399.115
	ATSTNUM	-0.24	0.395	-1.187	-0.608	0.546	-1.035	0.554	0.003	-0.086	-0.054	0.002	481.516
a Dependent Variable: SCAS.TOTAL													

Hierarchical Regression Predicting BBC with ACTIGRAPH Variables

Variables Entered/Removeda

Model	Variables Entered	Variables Removed	Method
1	PUBERTY, GENDER, AGEb	.	Enter
2	N.TOTALb	.	Enter
3	ATSTNUM, WASOMean, OnsetLatencyMean, meanwaketimeacti, meanBedtimeacti, actigraphawakeningsmean, EfficiencyMean, PercentSleepMean, ATIBNUMb	.	Enter

a Dependent Variable: BBC.TOTAL

b All requested variables entered.

Model Summaryd										
Model	R	R Square	Adjusted R Square	Std. Error of the Estimate	Change Statistics					Durbin-Watson
					R Square Change	F Change	df1	df2	Sig. F Change	
1	.074a	0.006	-0.045	12.861	0.006	0.11	3	59	0.954	
2	.632b	0.399	0.357	10.085	0.393	37.952	1	58	0	
3	.704c	0.495	0.361	10.055	0.096	1.038	9	49	0.424	2.039
a Predictors: (Constant), PUBERTY, GENDER, AGE										
b Predictors: (Constant), PUBERTY, GENDER, AGE, N.TOTAL										
c Predictors: (Constant), PUBERTY, GENDER, AGE, N.TOTAL, ATSTNUM, WASOMean, OnsetLatencyMean, meanwaketimeacti, meanBedtimeacti, actigraphawakeningsmean, EfficiencyMean, PercentSleepMean, ATIBNUM										

d Dependent Variable: BBC.TOTAL									

ANOVAa

Model		Sum of Squares	df	Mean Square	F	Sig.
1	Regression	54.342	3	18.114	0.11	.954b
	Residual	9759.086	59	165.408		
	Total	9813.429	62			
2	Regression	3914.379	4	978.595	9.622	.000c
	Residual	5899.05	58	101.708		
	Total	9813.429	62			
3	Regression	4859.265	13	373.79	3.697	.000d
	Residual	4954.163	49	101.105		
	Total	9813.429	62			

a Dependent Variable: BBC.TOTAL

b Predictors: (Constant), PUBERTY, GENDER, AGE

c Predictors: (Constant), PUBERTY, GENDER, AGE, N.TOTAL

d Predictors: (Constant), PUBERTY, GENDER, AGE, N.TOTAL, ATSTNUM, WASOMean, OnsetLatencyMean, meanwaketimeacti, meanBedtimeacti, actigraphawakeningsmean, EfficiencyMean, PercentSleepMean, ATIBNUM

Model		Unstandardized Coefficients		Standardized Coefficients	t	Sig.	95.0% Confidence Interval for B		Correlations			Collinearity Statistics	
		B	Std. Error	Beta			Lower Bound	Upper Bound	Zero-order	Partial	Part	Tolerance	VIF
1	(Constant)	66.072	16.675		3.962	0	32.706	99.437					
	AGE	-0.011	1.199	-0.001	-0.009	0.993	-2.41	2.389	-0.009	-0.001	-0.001	0.724	1.382
	GENDER	2.251	4.049	0.08	0.556	0.58	-5.852	10.354	0.071	0.072	0.072	0.813	1.231
	PUBERTY	-0.106	0.812	-0.022	-0.131	0.897	-1.732	1.52	0.009	-0.017	-0.017	0.613	1.632
2	(Constant)	83.694	13.385		6.253	0	56.902	110.486					
	AGE	-0.565	0.945	-0.072	-0.598	0.552	-2.455	1.326	-0.009	-0.078	-0.061	0.717	1.395
	GENDER	2.5	3.176	0.089	0.787	0.434	-3.857	8.857	0.071	0.103	0.08	0.813	1.231
	PUBERTY	0.18	0.639	0.037	0.281	0.78	-1.099	1.458	0.009	0.037	0.029	0.609	1.641
	N.TOTAL	-2.172	0.353	-0.631	-6.161	0	-2.878	-1.466	-0.621	-0.629	-0.627	0.988	1.012
3	(Constant)	-45.849	115.126		-0.398	0.692	-277.203	185.505					
	AGE	-1.582	1.231	-0.201	-1.285	0.205	-4.056	0.892	-0.009	-0.181	-0.13	0.42	2.383

	GENDER	2.081	3.489	0.074	0.596	0.554	-4.932	9.093	0.071	0.085	0.061	0.669	1.495
	PUBERTY	0.799	0.745	0.163	1.072	0.289	-0.699	2.297	0.009	0.151	0.109	0.445	2.248
	N.TOTAL	**-2.342**	**0.371**	**-0.68**	**-6.31**	**0**	**-3.087**	**-1.596**	**-0.621**	**-0.67**	**-0.641**	**0.887**	**1.128**
	MeanBedtime acti	-7.99E-05	0	-0.214	-1.574	0.122	0	0	-0.031	-0.219	-0.16	0.556	1.798
	Meanwaketi meacti	**-0.001**	**0**	**-0.381**	**-2.683**	**0.01**	**-0.001**	**0**	**-0.132**	**-0.358**	**-0.272**	**0.511**	**1.956**
	OnsetLatency Mean	0.238	0.241	0.429	0.986	0.329	-0.247	0.722	0.042	0.139	0.1	0.054	18.389
	EfficiencyMean	1.484	1.617	0.792	0.918	0.363	-1.766	4.734	-0.09	0.13	0.093	0.014	72.302
	WASOMean	0.329	0.229	0.505	1.434	0.158	-0.132	0.789	0.138	0.201	0.146	0.083	12.02
	PercentSleep Mean	0.228	2.177	0.063	0.105	0.917	-4.147	4.603	-0.116	0.015	0.011	0.029	34.831
	Actigrapha wakening smean	-0.144	0.148	-0.188	-0.972	0.336	-0.441	0.153	0.132	-0.138	-0.099	0.276	3.626
	ATIBNUM	0.306	0.288	2.153	1.062	0.294	-0.273	0.886	0.025	0.15	0.108	0.003	399.115
	ATSTNUM	-0.312	0.292	-2.381	-1.069	0.29	-0.899	0.275	0.012	-0.151	-0.108	0.002	481.516
a Dependent Variable: BBC.TOTAL													